CRANFIELD UNIVERSITY

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Evaluation of anaerobic digestion process for derived-MBT organic solid wastes

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Evaluation of anaerobic digestion process for derived-MBT organic solid wastes

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ABSTRACT

Semi-continuous and batch system bench-scale reactors, at mesophilic temperature, were set-up to investigate the performance of anaerobic digestion treating mixed waste from municipal solid collection, derived from two large-scale mechanical biological treatment (MBT) plant, in the UK.

The biogas yield using the semi-continuous reactor was determined to be between 300 and 410 mL/gVS with average methane content of 51% and average volatile solid destruction of 70%. During the batch system, the biogas yield was determined to be between 200 and 220 mL/gVS, with average methane content of 54% and volatile destruction of 70%, at the end of the digestion trial.

Heavy metal analysis indicated accumulation of some elements during the treatment; however, final values were lower than risk limits. Microbial community profile through phospholipid fatty acids (PLFA) analysis was investigated to determine structure shifts between different treatment and over time; although no statistical significance ($p>0.05$) was detected, PLFA peaks presented a shift in the community profile when comparing two different treatment applied. The community structure was demonstrated to be stable and resilient, able to cope with different parameters and stress.

The results of this study indicate that mixed municipal solid waste is favourable to be treated in anaerobic digestion plants with mechanical biological treatment (MBT) with regards to its biogas yield and waste size reduction through volatile solid destruction.

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“It's the end of the world as we know it and I feel fine” (R.E.M.)

EXECUTIVE SUMMARY

Until recently, almost all residual municipal waste in the UK was landfilled. The European Landfill Directive highlighted the need to reduce the biodegradable fraction of waste sent to landfill as DEFRA stated that in 2005 landfill contributed to 40% (928k tones) of the total UK methane emissions and that some of the landfill sites are still without methane control and collection.

A number of new waste treatment alternatives are being applied enabling the diversion of biodegradable waste from landfill site, such as composting, incineration, pyrolysis, gasification, anaerobic digestion and mechanical biological treatment (MBT). Anaerobic digestion is already well implemented on a widespread basis for the treatment of wastewater and source segregated streams, but still being adapted for other streams such as mixed fraction of the municipal solid waste, when the technology is applied in combination with MBT. Although the technology is already operational in large-scale in some European countries, it is essential to demonstrate and establish its efficiency in the UK.

The aim of the research was to evaluate the efficiency of anaerobic digestion technology when treating waste derived from two large-scale MBT plants, when submitted to different parameters. The set-up consisted of bench scale 5-litres chemostat reactors at semi-continuous loading system and 1-litre bottle reactors at batch loading system, both at mesophilic temperature. Trials consisted of different hydraulic retention time, loading rate and feedstock. The results of the trials, during semi-continuous system, demonstrated levels of biogas yield between 300 and 410 mL/gVS with average methane content of 51%; volatile solid destruction was between 60 and 70%. During batch system, the biogas

yield was determined to be between 200 and 220 mL/gVS, with average methane content of 54% and volatile destruction of 70%. There was no significant difference on the methane content after treatment with different parameters; however, the levels of H_2S increased significantly to over the limit of the equipment detection when the second waste stream was added.

Heavy metal analysis indicated accumulation of zinc, nickel and copper, during the treatment; however, final values were lower than risk limits. Microbial community profile through phospholipid fatty acids (PLFA) analysis was investigated to determine structure shifts between different treatment and over time; although no statistical significance was detected ($p>0.05$), PLFA peaks presented a shift in the community profile when comparing two different treatment applied. The community structure was demonstrated to be stable and resilient, able to cope with different parameters and perturbations.

The experiment proved anaerobic digestion with mechanical biological system treating municipal solid waste, to be an efficient alternative to landfill when applied to mixed waste streams, with considerable biogas yield. Although the quantity and quality of the biogas was lower when compared to anaerobic digestion of source segregated waste, the technology could still be an important asset in an integrated waste management strategy, producing renewable energy, as a promising alternative to landfill.

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ABBREVIATIONS

AD: anaerobic digestion.

BAT: best available technique

BSE: bovine spongiform encephalopathy.

BMW: biodegradable municipal waste, also called organic fraction of municipal waste (OFMSW); is the fraction capable of undergoing anaerobic or aerobic decomposition.

CH_4: methane.

C:N: carbon to nitrogen ratio.

CO_2: carbon dioxide.

COD: the quantity of oxygen used in biological and non-biological oxidation of materials in water; a measure of water quality.

CSTR: continuous stirred tank reactor.

DEFRA: Department for Environment, Food and Rural Affairs.

EA: Environment Agency.

EU: European Union.

FAME: fatty acid methyl ester.

GHG: greenhouse gas.

HRT: hydraulic retention time; reactor volume / flow rate.

H_2S: hydrogen sulfide (or sulphide).

ICP-MS: Inductively coupled plasma mass spectrometry.

IPCC: Intergovernmental Panel on Climate Change.

L: litre - although not an SI unit, it is accepted for use with the SI.

LA: local authority.

LATS: Landfill Allowances and Trading Scheme; LAS in Scotland.

LPG: liquefied petroleum gas.

MBT: mechanical biological treatment.

MSW: municipal solid waste.

OFMSW: see BMW

OLR: organic loading rate.

RCF: relative centrifugal force.

SEPA: Scottish Environment Protection Agency.

SRF: solid recovered fuel.

PLFA: phospholipid fatty acid.

PAS 100: Publicly Available Specification 100.

ppm: parts per million.

SI: Statutory Instruments.

SR: Statutory Rule (Northern Ireland).

SSI: Scottish Statutory Instruments.

Tpa: tonnes per annum.

TSE: transmissible spongiform encephalopathy.

UNFFC: United Nations Framework Convention on Climate Change.

v/v: "by volume"; used to describe the concentration of a substance in a mixture or solution (i.e. 2% v/v = the volume of the substance is 2% of the total volume of the solution or mixture).

WSI: Welsh Statutory Instruments.

CHAPTER 1

INTRODUCTION

1.1 Background of the research

The increase of greenhouse gases (GHG) concentrations over the last 100 years has caused the temperature of the Earth to rise by 0.6 ˚C (Eurostat, 2005). Carbon dioxide (CO_2) and methane (CH_4) are among the major GHGs (Figure 1.1) present in the atmosphere (Houghton *et al.*, 2001; SSEFRA, 2006) and are thus subject to the Kyoto Protocol, which aims to achieve the "stabilization of greenhouse gas concentrations in the atmosphere at a level that would prevent dangerous anthropogenic interference with the climate system" (UNFFC, 1997). Under the Kyoto commitments, the United Kingdom agreed to reduce its greenhouse gases emissions by 12.5% below 1990 levels (base year) in 2008-2012 (SSEFRA, 2006).

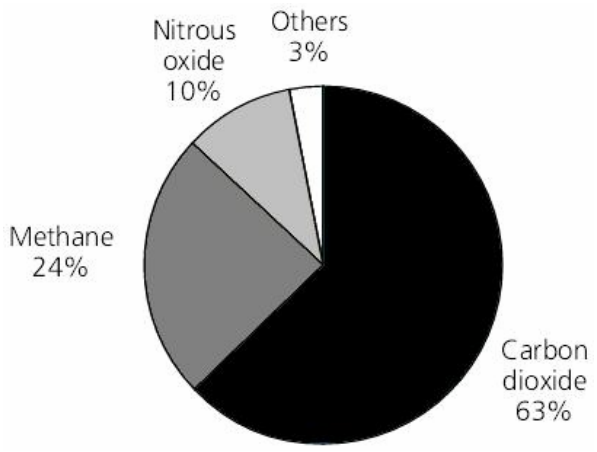

Figure 1.1 – The relative contribution to global warming over the next 100 years of current emissions of greenhouse gases (SSEFRA, 2006).

The degradation of organic waste (e.g. kitchen waste, garden waste, paper) by microorganisms can produce carbon dioxide and methane and adequate treatment and disposal of such materials have a direct influence on the emissions of these gases (DEFRA, 2005). In the UK landfill is the main waste disposal method; approximately 70% of the waste is being buried (Figures 1.2-1.3), and some of the landfill sites are still without methane collection (SEPA, 2007). It has been estimated (Eurostat, 2005) in the UK, that an average of 375 kg of municipal waste per person is sent to landfill annually (Figure 1.4), where 40% (928k tones) of the UK total methane emissions (Figure 1.5) are produced (DEFRA, 2005).

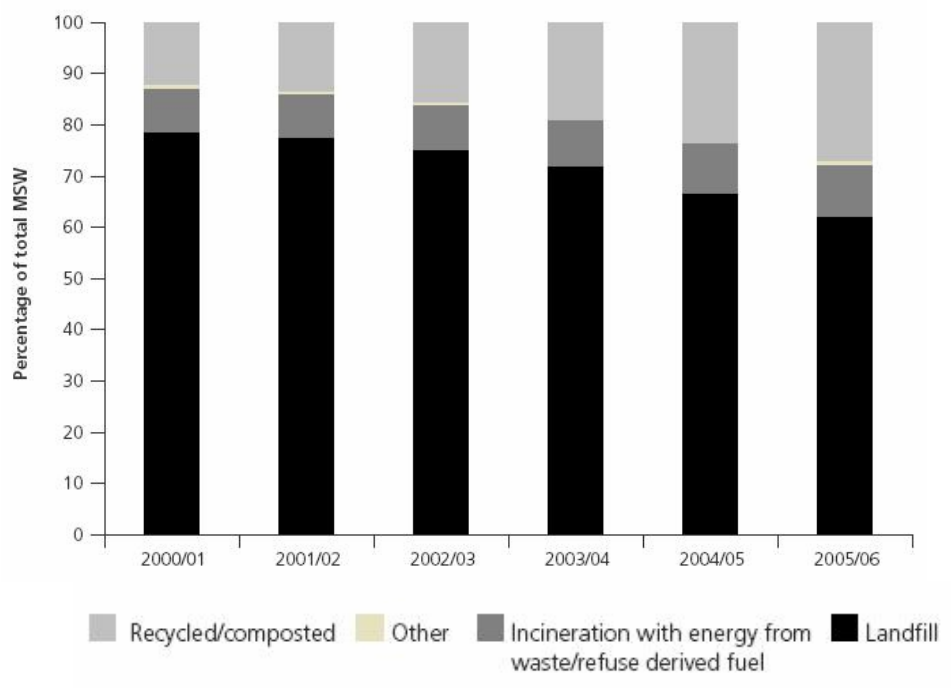

Figure 1.2 – Management of municipal waste in England, 2000/2006 (DEFRA, 2007).

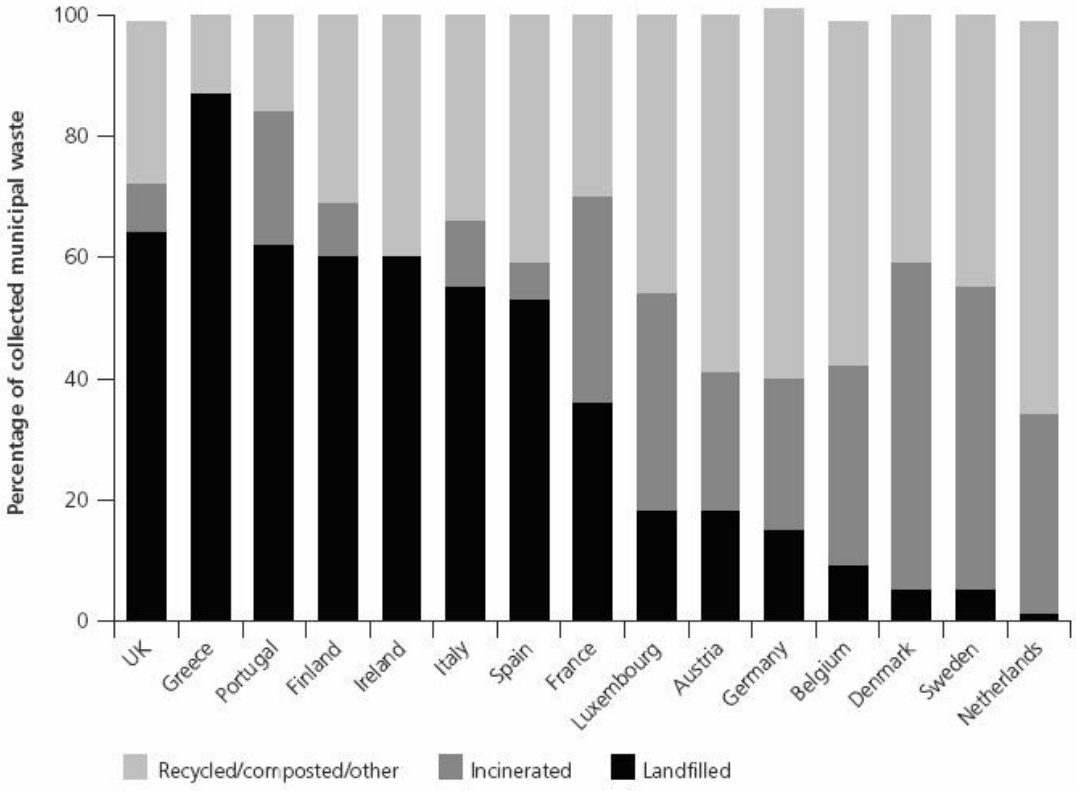

Figure 1.3 – Management of municipal waste within EU members (Eurostat, 2005).

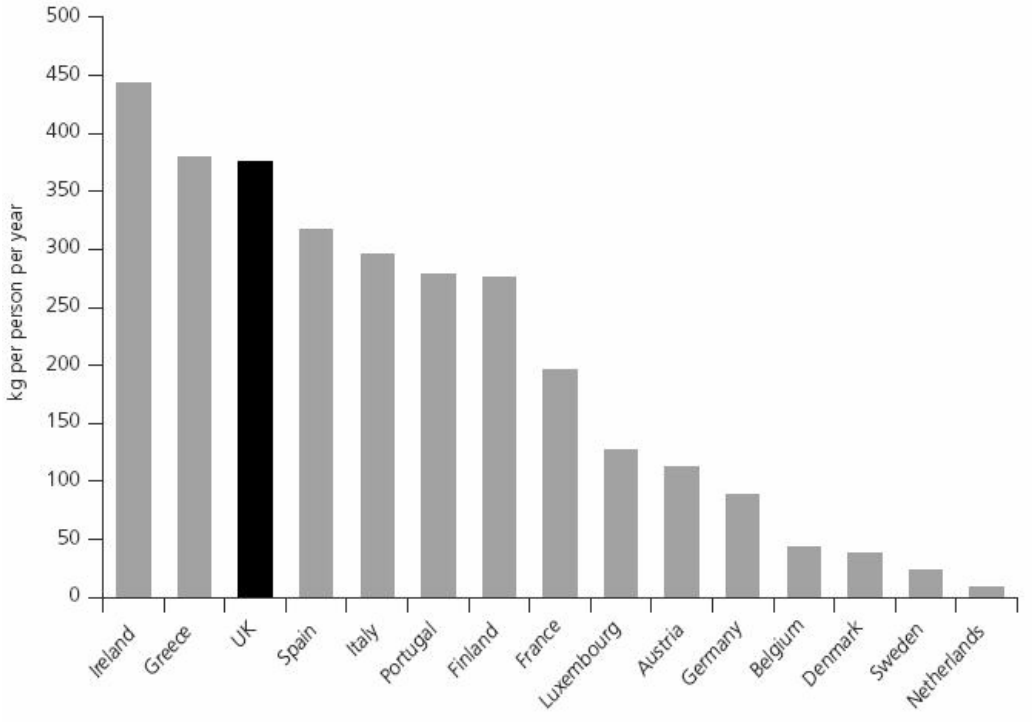

Figure 1.4 – Municipal waste sent to landfill within EU members (Eurostat, 2005).

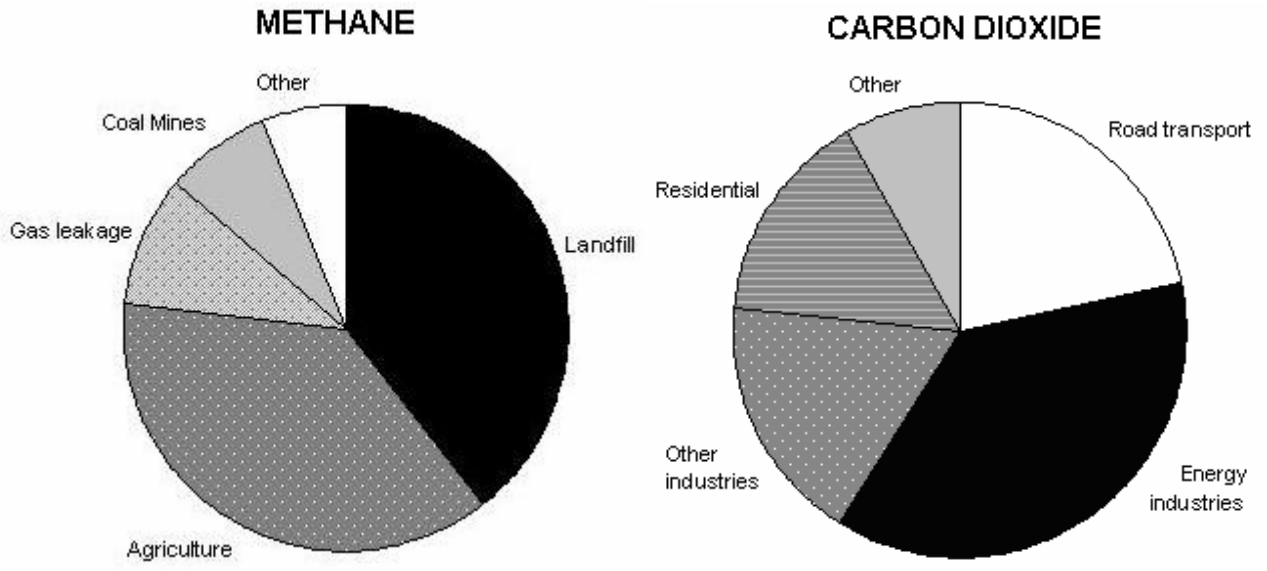

Figure 1.5 – Methane and carbon dioxide emissions by source in the UK, 2005 (DEFRA, 2007).

The UK reliance on landfills can be explained by the historical abundance and low cost of sites, previously used for quarrying and mining. Nevertheless, landfill space is running out and gate fees are increasing while waste generation is also increasing (DEFRA, 2007). As a consequence, emphasis is now being placed on alternative waste treatment.

In 2005, about 42% of UK renewable gas and electricity were produced from landfill gas (DEFRA, 2005). Although the carbon emissions produced from the combustion of landfill biogas do not count towards national emissions, burying materials reduces the potential for recycling, reusing and recovering resources and increasing consumptions of raw resources (DEFRA, 2005).

The EU Landfill Directive (1999/31/EC) set reduction targets on the amount of biodegradable wastes sent to landfill, requiring alternative waste management practices. One guiding principle adopted by UK and European waste management practices options was the concept of waste hierarchy (Figure 1.6), where the most desirable option is the waste prevention and the least suitable method is disposal without recovery of materials or

energy. The waste hierarchy has influenced the designing of national policies and plans to move the UK away from its dependence on landfill (SITA, 2004).

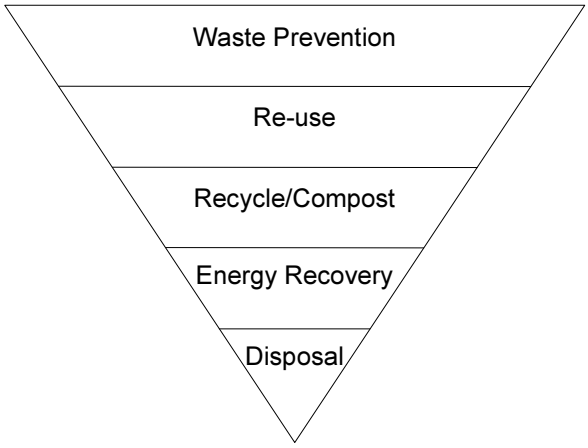

Figure1.6 – Waste hierarchy (DEFRA, 2007).

A number of alternative technologies are expected to receive the diverted waste (e.g. anaerobic digestion, mechanical biological treatment, composting, incineration, pyrolysis and gasification); amongst those, anaerobic digestion has been favourably recommended in the recently published Waste Strategy for England (DEFRA, 2007)

In several European countries large-scale mechanical biological treatment (MBT) with anaerobic digestion plants have been operating to produce renewable energy as well as allowing recovery of recyclable material from waste not adequately segregated at source. This technology has not been widely adopted in the UK and perceived barriers should be further investigated to inform the design and operation of the systems. There is also limited literature investigating the treatment of waste derived from MBT with anaerobic digestion, for waste streams from UK.

1.2 Aims and Objectives

The aim of the research was to evaluate the efficiency of anaerobic digestion technology in treating waste derived from mechanical biological treatment (MBT), as well as understand and assess which parameters are influencing the quality of biogas production in a laboratory scale digester.

In order to achieve these aims, the following objectives were proposed:

- To evaluate and assess the volume and profile of biogas production as well as parameters influencing it;
- To identify the influence of the waste characteristics on the process performance.

1.3 Expected outcomes

Due to the lack of a broad literature available on the treating of waste derived from mechanical biological treatment with anaerobic digestion for waste streams from the UK, the findings of the present research expects to provide a base-knowledge to implement anaerobic digestion as a waste treatment technology when combined with mechanical biological treatment.

CHAPTER 2

ANAEROBIC DIGESTION

2.1 Background of the process

Anaerobic digestion (AD) is a process in which organic matter is decomposed by microorganisms in absence of oxygen. The chemistry of the process is the same as that found in landfills, the difference being that digestion occurs in enclosed and controlled systems (Williams, 2005) and over a shorter period of time (McDougall *et al*., 2001).

A simplified overall chemical reaction of the process can be summarised as (Evans, 2001; IWM, 1998; Polprasert, 1996):

$$\text{Organic matter} \xrightarrow{\text{Anaerobic Digestion}} CH_4 + CO_2 + H_2 + NH_3 + H_2S$$

Edelmann *et al.* (2000), after life cycle and economical analysis, reported anaerobic digestion as the most advantageous waste treatment due to its better energy balance, when compared to incineration, composting and a combination of composting and digestion. The analysis also revealed that methane emissions in compost treatment were considerable. Björklund *et al.* (1999) reported anaerobic digestion with reduced environmental impact when compared composting, using life cycle analysis. Mata-Alvarez *et al*. (1993) and Vallini *et al*. (1993) also demonstrated the anaerobic digestion process as the best option in terms of energy and mass balance.

Anaerobic digestion presents several environmental benefits. It contributes to the reduction of GHG emissions to atmosphere with diversion of disposal of organic waste in

landfills. It allows the production of renewable energy (heat, electricity and transport fuel) from the biogas and a nutrient rich digestate, as a renewable alternative to mineral fertilisers.

2.2 Anaerobic digestion stages

The degradation of organic matter in the anaerobic digestion process takes place through four key stages (Figure 2.1): hydrolysis, acidogenesis, acetogenesis and methanogenesis (Braber, 1995; Dearman *et al*., 2006). These stages result from the biological activities of two key bacterial groups: acetogens and methanogens. Chemical reactions in one stage affect the following stage (Wheeler and Rome, 2002) in a syntrophic and mutualistic relationship (Dearman *et al.*, 2006).

2.2.1 Hydrolysis

At the first stage of the process, polymers of carbohydrates, lipids and proteins are converted into soluble organic compounds by enzymes secreted by hydrolytic bacteria (Garcia *et al*., 2000; Wheeler and Rome, 2002). Proteins of the waste are converted into amino acids, fats into long-chain fatty acids, and carbohydrates into simple sugars (Kaseng *et al*., 1992; Monnet, 2003; Parawira *et al*., 2005).

This step is the rate-limiting of the digestion process and depends on the substrate availability, bacterial population density, temperature and pH (Gayat, 2002). Waste containing lignin (such as woody waste) due to its complex polymers units, can slow down the process since intact it is biodegraded slowly (Eriksson, 1990; Maier *et al.,* 2000).

2.2.2 Acidogenesis

At the second stage of the process, soluble organic compounds, including the products of the hydrolysis, are fermented into various intermediate products such as short chain organic acids, volatile fatty acids (VFA) and alcohols such as methanol (Wheeler and Rome, 2002). The main products of this stage are acetic acid, lactic acid and fatty acid and the pH falls as the levels of these components increase. Production of energy is relatively low and the reduction in the organic load is minimal.

2.2.3 Acetogenesis

This is an important step in the overall process since it involves the breakdown of alcohol and fatty acids into acetic acids, carbon dioxide and hydrogen by acetogens bacteria (Equation 2.1), such as *Desulfovibrio* spp., & *Desulfomarculum* spp. (sulfate-reducing bacteria); *Syntrophobacter* spp. and *Syntrophomonas* spp. (Wheeler and Rome, 2002; Gerardi, 2003). The hydrogen has a critical role in the digestion as under standard conditions, it inhibits oxidation. Therefore, hydrogen-scavenging bacteria are required to ensure the conversion of all acids, lowering partial pressure. As a result, the concentration of hydrogen, measured by partial pressure, is an indicator of the health of a digester (Mata-Alvarez, 2003).

$$CH_3CH_2OH + CO_2 \rightarrow CH_3COOH + 2H_2 \qquad (2.1)$$

2.2.4 Methanogenesis

The methanogens (also called methane-producing bacteria) such as *Methanobacterium* spp.; *Methanosarcina* spp.; *Methanococcus* spp.; *Methanothrix* spp., are the final components of the anaerobic food chain (Gerardi, 2003). Their metabolic activity prevents the sequestering of large quantities of organic material in anaerobic ecosystem by converting products of the previous stage like acetate (acetotrophic or aceticlastic bacteria) and hydrogen (hydrogenotrophic bacteria) to methane and carbon dioxide (Equations 2.2 and 2.3) (Gerardi, 2003; Stanier *et al.*, 1989; Wheeler and Rome, 2002). According to Klass (1984) 70% of the methane production came from the acetate, whilst the remainder is originated from H_2 and CO_2. The presence of ammonia, produced from the degradation of nitrogen-rich protein such as those in the blood, is toxic to methanogenic population and can cause process failure (Banks and Wang, 1999; Wang and Banks, 2003).

Sulfate-reducing bacteria reduce sulfate to hydrogen sulphide (H_2S), during degradation of organic compounds, using hydrogen, which is also competed by methanogens. The hydrogen sulfide produced has inhibitory effect on methanogens even at low concentrations (Gerardi, 2003). Hydrogen sulphide has also toxic effect in humans when inhaled, suppressing aerobic metabolism by inhibiting cytochrome oxidase (Bhambhani and Singh, 1991).

$$CH_3COOH \rightarrow CH_4 + CO_2 \qquad (2.2)$$

$$CO_2 + 4H_2 \rightarrow CH_4 + 2H_2O \qquad (2.3)$$

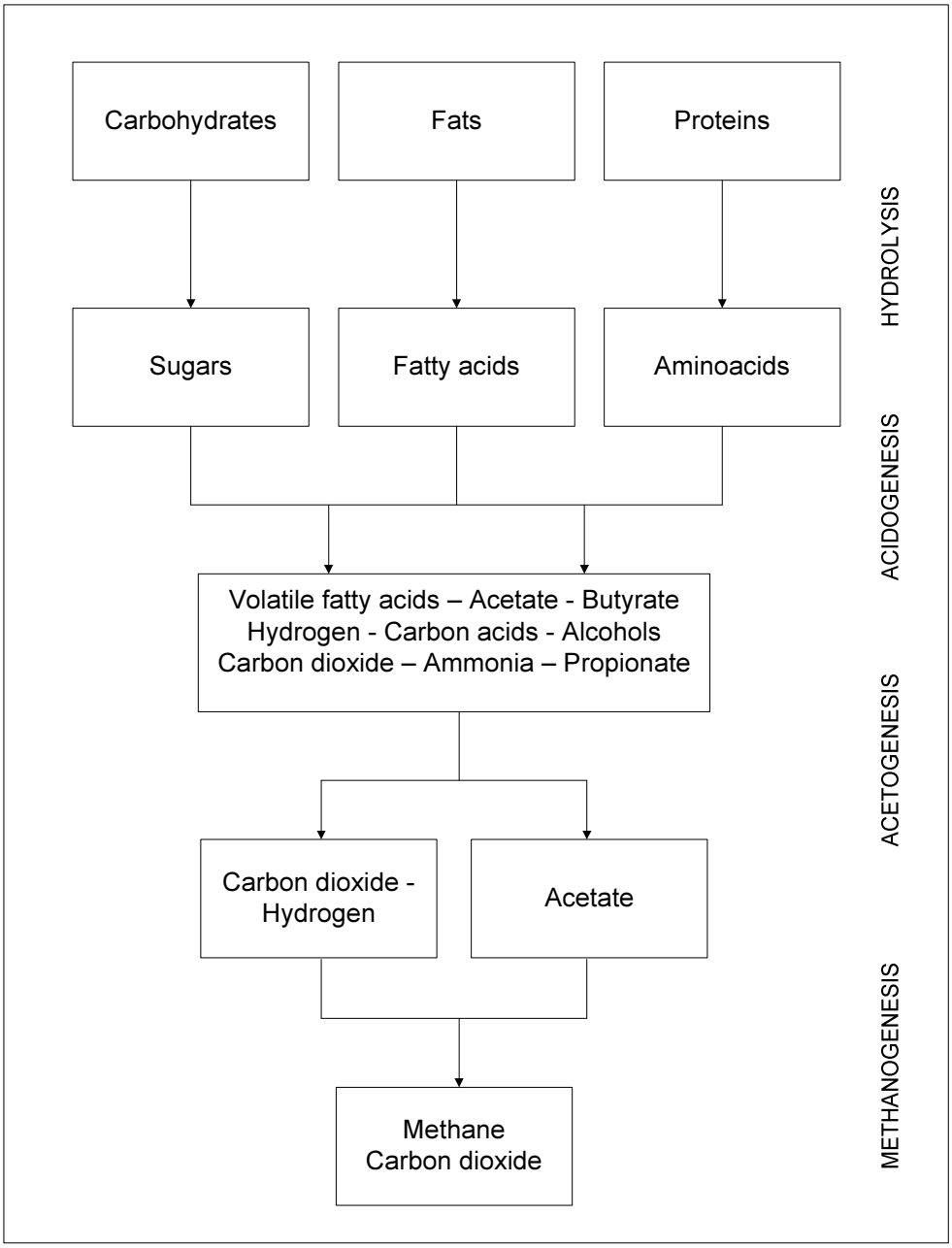

Figure 2.1 – Anaerobic digestion stages (Gujer and Zehnder, 1983; Siegrist *et al.*, 1993)

2.3 Operational conditions

2.3.1 Start-up of anaerobic digestion

Prior any digestion to occur, it is necessary to initialise and stabilise the system in order to achieve an optimum temperature and bacteria content. The start up process can be placed for a new digester or for restarting a sour one and it can be in a ‘natural’ or ‘pH control’ process (Toprak, 2006). The tank is filled with inoculum/seed from manure or sludge treatment plant (Fruteau, 2006; Nordberg, 2006) or fresh wastewater (to reduce the start up time) and heated to the desirable temperature and maintained (Barker, 2001). The feedstock material should be added gradually and increased over a period of weeks to the desired loading rate (Barker, 2001; Fruteau, 2006; Toprak, 2006). The recirculation, the control and composition of the biogas, and control of volatile fatty acids, pH, dry solids and ammonia must be done on a daily basis, until the designed load is reached (Fruteau, 2006; Potts and Jolly, n.d; Toprak, 2006).

2.3.2 Feedstock

The first and foremost issue when considering the implementation of anaerobic digestion systems is the feedstock. The term feedstock is defined as any raw material used in an industrial process; in the AD context, it is considered to be any substrate that can be converted to methane (Steffen *et al.*, 1998). Thanh (1982) reported that all organic materials, except mineral oil and lignin, are suitable feedstock for anaerobic digestion plants.

Choice of the feedstock reflects on the behaviour of the process and also interferes on the quality of the outputs (Steffen *et al.*, 1998). As an example, lignin degradation is difficult

and slowly while cellulose degradation can occur in several weeks; on the other hand, hemicellulose, protein and fat are degraded within few days, while volatile acids, alcohol and sugar are degraded in hours. Comparing to carbohydrates and protein, fat is the substance that provide highest biogas yield but requires the highest retention time (Steffen *et al.*, 1998). The moisture content is also an important factor related to feedstock: the wetter the material the more suitable the material will be to handling within pumps instead of screw presses and physical means of movement, and it will also takes more space and volume.

2.3.3 Co-digestion

Co-digestion is the use of co-substrates to improve the biogas yields in a synergism interaction, supplying the missing nutrients. The most common co-digestion is between biodegradable municipal waste and organic waste as sewage sludge (Mata-Alvarez *et al*. 2000). However, digestion of mixture of different wastes is seldom reported (Carucci *et al.*, 2005; De Baere, 2000).

2.3.4 Temperature requirements

The natural incidence of methanogens demonstrates that anaerobic degradation can occur with temperatures ranging from 10 ℃ to over 100 ℃ , and at a moisture contents ranging from 60% to more than 99% (Wheeler, 2001).

Bacteria are usually divided into several classes based upon temperature optima. The low temperature bacteria are the psychrophiles, which can grow at temperatures down to -10 ℃, but whose optimum temperature is 15 ℃ or lower . The mesophiles thrive at medium

temperatures, 20-45 ℃, and include human pathogens . Thermophiles thrive above 45 ℃, while hyperthermophiles live from 65 ℃ or even abo ve the boiling point of water (Gerardi, 2003).

There are two conventional operational temperature levels for anaerobic digesters, which are determined by the community of methanogens in the reactors (Figure 2.2):

- Mesophilic temperature: which takes place optimally around 35 ℃;
- Thermophilic temperature: which takes place optimally around 55 ℃.

The advantage of mesophilic operation is that it is well understood, requires less heat to maintain operation, achieves a greater degree of stabilisation and is more robust due to the larger diversity of mesophiles in nature (Wheeler and Rome, 2002; Gerardi, 2003). However, the gas yield is lower and, if sanitisation is required, this must be conducted in a separate stage. The retention time, period in which the feedstock remains in the digester, is typically between 15 and 30 days (British Biogen, n.d.).

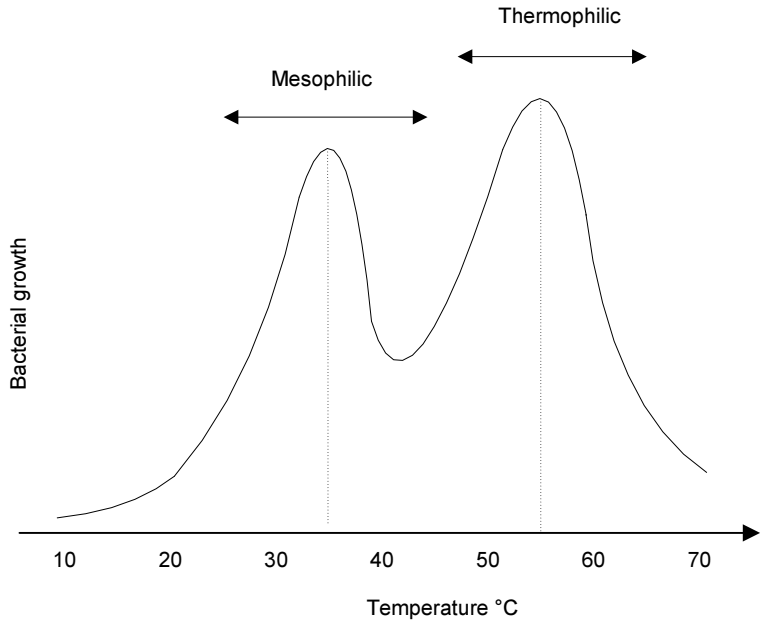

Figure 2.2 – Temperature ranges used for anaerobic digestion with optima for mesophilic and thermophilic (Mata-Alvarez, 2003).

Thermophilic process operates at a faster rate with bacterial growth rate 2 to 3 times higher than mesophilic (Zábranská *et al*., 2000; Mladenovska and Ahring, 2001) and achieves complete and faster sanitisation of the waste (Lo *et al*., 1985; Wheeler and Rome, 2002; Zábranská *et al*., 2000; Ahring *et al*., 2001; Olsen and Larsen, 1987). The retention time ranges from 12 to 14 days (British Biogen, n.d.; Gerardi, 2003) and produces higher methane yield (British Biogen, n.d.).

Nevertheless, in many cases, the additional energy input necessary for the thermophilic process is approximately the same amount produced in these conditions (Mata-Alvarez *et al*., 2000). The thermophilic process also requires a higher degree of operation and monitoring: the number of thermophiles is very limited and the bacteria are sensitive to temperatures fluctuation.

Some bacteria can grow in wider range of temperatures from 22 ℃ to 78 ℃ (Wiegel, 1990). However, the change of temperature from mesophilic to thermophilic conditions needs time for the adaptation of anaerobic biomass and for the selection of thermotolerants (Zábranská *et al*., 2000).

2.4 Parameters that influence anaerobic digestion

As anaerobic digestion involves a complex and diversified group of microorganisms, monitoring is required to determine any irregular behaviour in order to take actions in the early stages, preventing system failure (Table 2.1). Among the parameters that indicates the stability of the digester are: COD, volatile solids removal (conversion/removal); volatile fatty acids, pH, alkalinity, H_2, CO (accumulation); CH_4, CO_2 (outputs) and microbial activity.

2.4.1 Biogas production

The biogas production is an important and the most common parameter to be measured. As the production depends on loading rate and feedstock composition, it cannot be used to indicate process imbalance and it is not effective as an early warming parameter (Moletta *et al.,* 1994; Switzenbaum *et al*., 1990). According to Liu *et al.* (2004) the methane production is a better parameter than biogas composition.

2.4.2 pH

The pH is an important and easily to measure parameter to demonstrate digester good performance (Dearman *et al*., 2006). According to Golueke (2002) the ideal pH for the bacteria in the process is around 5.5 to 8.5, but the optimum for methanogenesis is close to neutral. The low pH indicates an acid accumulation – volatile fatty acids – from the hydrolysis stage and can cause system failure. The only parameter that shows instability faster than pH is gas production (Golueke, 2002; Monnet, 2003).

2.4.3 Temperature

Temperature control is important to maintain the required range in the process (mesophilic or thermophilic). Temperature variation during the process may affect gas production since anaerobic bacteria are heat sensitive (Monnet, 2003).

2.4.4 Carbon:Nitrogen ratio (C:N)

According to Monnet (2003) the ideal C:N-ratio is around 25 while for Steffen *et al.* (1998) the optimum is a C:N:P-ratio of 100:5:1. High levels of nitrogen (meaning a low ratio) can cause excess of ammonia that is toxic for methanogens. On the other hand low levels of nitrogen (high ratio) will be rapidly consumed by methanogens resulting in a decrease of gas production (Monnet, 2003).

2.4.5 Retention time

Retention or residence time is related to the time required for the waste to remain in the digester tanks to complete the organic degradation and it is measured by BOD and COD (Ostrem, 2004). The appropriate time will depend on the quality of the feedstock and environmental factors, but for dry process it ranges between 14 and 30 days. Mixing is a practice that helps to reduce the retention time since it avoid the settlement of residues at the bottom of the tank (Vlyssides and Karlis, 2004).

2.4.6 Organic loading rate

Organic loading rate (OLR) is a measure of the biological conversion capacity. A high content of organic material will stimulate growth of acidogenic bacteria and elevated production of acid compounds as a consequence. The surplus of acid may cause system failure. The indication of this parameter is the low production of gas and a low pH (Mata-Alvarez, 2003; Monnet, 2003).

2.4.7 Volatile fatty acids

Volatile fatty acids (VFA) is considered an important early warning of the process failure (Ahring *et al*., 1995; Hill and Holmberg, 1988; Hill and Bolte, 1989). VFA such as propionic and butyric acids formed from degradation of molecules like proteins, fats and carbohydrates are the main substrate for the last two stages of digestion. Fast degradation increase the VFA levels, and combined with low pH values can cause system failure (Mackie and Bryant, 1995; Mata-Alvarez, 2003).

2.4.8 Hydrogen

Hydrogen accumulation is considered an early stage indicator for the process instability since high concentrations can inhibit volatile acids degradation, resulting in VFA accumulation (Collins and Paskins, 1987; Steyer *et al*., 2002). The presence of compounds such as sulfur, ammonia, xenobiotics substances (Mata-Alvarez, 2003) and high contents of potassium and lipids (Carucci *et al*., 2005) are also inhibitory or toxic to the anaerobic environment.

2.4.9 System improvement

The performance of anaerobic digestion can be improved to produce outputs with higher quality and quantity. Sonakya *et al.* (2001) observed that the H_2 production could be decreased 28% or increased 152% and methane production could be increased up to 110% as a function of different microorganisms (e.g. *Bacillus subtilis*, *B. licheniformis* and *Aspergillus niger*) and hydrolytic enzymes, individually and in combinations. The addition

of hemicellulose-degrading bacterium B4, could also result in a 30% increase in the biogas production (Mata-Alvarez *et al.*, 2000).

Table 2.1 – Possible disturbances of anaerobic digester and predictable results (Mata-Alvarez, 2003).

Disturbance	Arising Problem	Final Effect if not Digester Total Failure
Flow rate increase	Washout of microorganisms. Methanogens are the most affected, given its doubling time	Reduction in: methane % in gas pH methane production rate Alkalinity
Feed concentration increase (overloading)	Imbalances mainly affecting methanogenic bacteria and resulting in an accumulation of VFAs	Increase in: VFAs Acids different from acetic acid
Introduction of toxic substances		
Temperature fluctuations Oxygen exposure		

2.5 Types of digesters

The anaerobic digestion process can be processed in batches or continuously, and in one or two-phases. The digesters can also be classified as covered lagoons; complete stirred tanks; anaerobic filter reactors; upflow sludge blankets and fluid bed reactors.

2.5.1 Batch system

In the batch system, the biomass is added to the reactor at the beginning and sealed for the duration of the process. The biogas production occurs in a normal distribution pattern – which can be used as a monitoring parameter. The retention time can take from 2 to 3 weeks. After the digestion, the reactor is opened and the effluent is removed without contact with the new load (Braber, 1995).

Compared to continuous system, batch is simpler, less expensive and more robust; nevertheless, the gas yield can be lower. Batch reactors can also suffer from risk of explosion during emptying cycles (Monnet, 2003; Vandevivere *et al*., 2003).

According to Vandevivere *et al.* (2003) the batch system can be compared to a “landfill-in-a-box”; however, the production of biogas can achieve 50 to 100 fold higher than that of a landfill; the reason is the use of recirculation and higher temperatures in the anaerobic digestion (Lissens *et al*., 2001; Vandevivere *et al*., 2003; Wheeler, 2001).

2.5.2 Continuous system

In the continuous system, which is commonest type, organic matter is constantly added, or added in stages to the reactor, while the end products are removed, resulting in constant production of biogas. Here, new materials are mixed with the liquid remaining in the tank (Ostrem, 2004).

2.5.3 ‘Dry’ and ‘wet’ mode

‘Dry’ mode digesters have a high solid content - 20 to 40% total solid, while ‘wet’ mode have a low solid content - 10 to 15% (Lissens *et al.*, 2001; Vandevivere *et al.* 2003); if total solid is higher than 50% it needs to be diluted (Oleszkiewicz and Poggi-Varaldo, 1997). ‘Dry’ mode produce thick slurry that will require more energy input to move and process the feedstock. It has a lower land requirement due to the lower volumes associated with the moisture.

‘Wet’ mode can transport material through the system using pumps that require significantly lower energy input. It requires a larger amount of land than high solids due to increased volumes. There are benefits associated with operation in a liquid environment such as enabling more thorough circulation of materials and contact between the bacteria and the substrate.

‘Wet’ mode was the main process adopted during the 1980’s while in the 1990’s, the erected plants were evenly split between the two modes (De Baere, 2000). Although the initial challenge in the ‘dry’ mode was related to handling, pumping and mixing the streams, this system has already proven reliable in France and Germany for the treatment of mechanically sorted biological municipal waste (Vandevivere *et al*., 2003).

2.5.4 One and two-phase process

In the one-phase (one-stage or one-step) process, the digestion occurs in one reactor tank and in ‘dry’ or ‘wet’ mode (Lissens *et al.*, 2001; Vandevivere *et al.* 2003). Two-phase process is used to separate de hydrolysis from the acetogenesis and methanogenesis stages, as the biochemical reactions do not necessarily share the same environmental

conditions, therefore different designs can be applied, as a combination of meso and thermophilic temperatures (Vandevivere *et al.* 2003). The first stage rate is limited by composition of the waste such as lignocellulose compounds and the second is limited by methanogens bacteria growth (Banks and Wang, 1999; Lissens *et al*., 2001; Liu and Ghosh, 1997; Palmowski and Müller, 2000). To avoid clogging in the filters, solids must be removed and methanogenic bacteria are retained in trapping devices, so that they are not flushed out (Braber, 1995).

The separation of the process in two phases enables the individual control of each stage with increased biological stability; performance and the biogas yield are also higher (Lissens *et al*., 2001). Research by Pavan *et al.* (2000) with source segregated waste suggested the two-phase system as compulsory for highly biodegradable wastes. According to Weiland (1993), the main advantage of the two-phase system is not necessarily reflected in higher yields but greater biological reliability for wastes which cause unstable performance (Table 2.2).

Although two-phase systems are preferred in laboratory analysis, since it is easier to control each step (Vandevivere *et al*., 2003), De Baere (2000) argued that its benefits are not yet proved and high digestion rates have been obtained in one phase system.

Table 2.2 – Advantages and disadvantages of two-phase systems (Vandevivere *et al.* 2003).

Criteria	Advantages	Disadvantages
Technical	Design flexibility	Complex
Biological	More reliable for cellulose-poor kitchen waste Only reliable design (with biomass retention) for C:N<20	Smaller biogas yield (when solid not methanogenized)
Economic & Environmental	Less heavy metal in compost (when solid not methanogenized)	Larger investment

2.6 Anaerobic digestion with mechanical-biological treatment

Mechanical-biological treatment (MBT) as a waste treatment technology was developed in Germany (*mechanisch-biologische abfallbehandlungsanlagen*) in the 90's to enable resource recovery from unsorted or sorted municipal solid waste streams (Archer *et al.*, 2005). The process is not regarded as a final disposal option but a mixture of integrated processing operations (Williams, 2005). MBT is a general term that includes several technologies applied at both mechanical (e.g. trommel, magnet current, hammer mill, Eddy current, shredders, hand picking) and biological stages (e.g. windrow or in-vessel aerobic composting, bio-drying, anaerobic digestion) (Archer *et al.*, 2005; Soyez and Plickert, 2002).

Variations of the technologies applied during the treatment and the order of these lead to different terminologies. As an example 'biological-mechanical treatment' (BMT) has the biological stage prior to mechanical while 'biological-mechanical stabilisation' (BMS) has a

stabilisation step at the end (Archer *et al.*, 2005). Other terminology applied is mechanical-biological pre-treatment (MBP), considering the technology as not a final disposal option. The term MBT was adopted in this document as a general mechanical stage followed by an anaerobic digestion process, unless otherwise stated.

The MBT technology is already implemented in European countries such as Germany, Belgium, France, Italy and Spain (Archer *et al.*, 2005; Williams, 2005), in fully commercial (i.e. at least two plants operating for more than one year) or commercial plants (i.e. one plant operating for more than one year). The capacity scale of these varies from 20,000 to 200,000 tonnes per annum (Archer *et al.*, 2005; McLanaghan, 2002). Worldwide it is also present in pilot plants (Bezama *et al*., 2007; Defra, 2005; Münnich *et al*., 2006; Soyez and Plickert, 2002).

The main objective of the mechanical stage is to maximise resource recovery (e.g: glass, plastic and metals) as well as fractionate and homogenise particle size to optimise its biodegradability during the biological stage (Figures 2.3-2.4) (Archer *et al*., 2005; Soyez and Plickert, 2002). The biological stage is selected according to the type of output material required (e.g. biogas, solid recovered fuel, fully bio-stabilised solid); the quantity of waste to be treated; regulatory requirements and economical/technical/commercial factors (Archer *et al.,* 2005). Other aspects that influence the process design are the minimisation of the waste biodegradability; outputs complying with market requirement; minimisation of environmental impacts and visual profile of the facility (Archer *et al.*, 2005.).

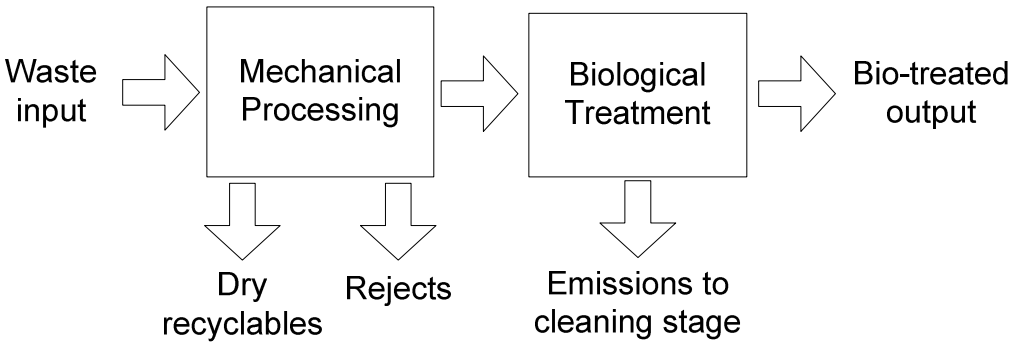

Figure 2.3 – General MBT diagram.

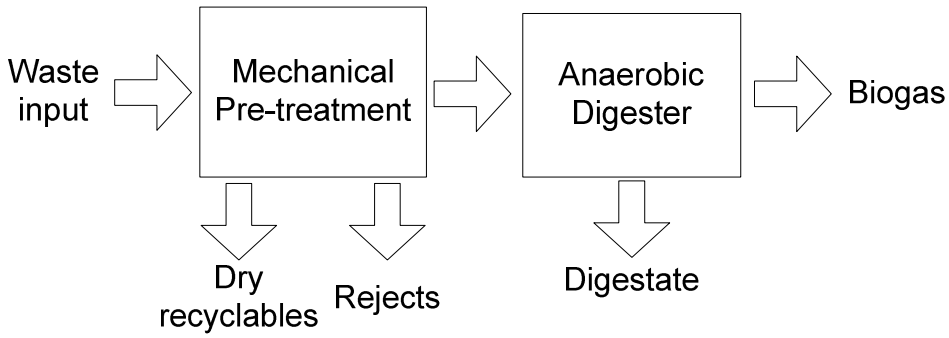

Figure 2.4 – MBT with anaerobic digestion.

Considering the advantages of the process, MBT demonstrate significant potential as it can achieve landfill diversion targets; increase recycling performance; increase sustainability of waste management; benefit from positive public perception when compared to thermal treatments; demonstrate lower technology risk associated, with well proven systems available, when compared to other novel approaches (Archer *et al.*, 2005; McLanaghan, 2002; Robinson *et al.*, 2005). The process also allows the final treated waste to be landfilled, when more sustainable options are not applied, as stabilised material (Muller *et al.*, 1998; Stegmann, 1996).

2.6.1 Municipal solid waste

The municipal solid waste (MSW), by its definition, is the fraction of waste composed by household waste, commercial waste and institutional waste, collected by or in behalf of a local authority (Williams, 2005). MSW, due to its constituents, presents diversified physic-chemical characteristics, what represents a difficulty when choosing the best treatment option. The waste can be comprised of paper and cardboard, food and garden waste, plastics, ferrous and non-ferrous metals, glass, textiles, and other minor fractions, and its composition can vary according to seasonal, geographic and socio-economic conditions (Williams, 2005).

The organic fraction of municipal solid waste (OFMSW) is the fraction that will breakdown under biological process and is the main feedstock for anaerobic digestion process. It can be classified as unsorted collection (all materials being placed together in one container and further sorted mechanically – MS-OFMSW), and separate collection, which can be split in two categories: separately collected from restaurants and canteens (SC-OFMSW) and domestic/household source sorting (SS-OFMSW) (Cecchi *et al.*, 2003). A comparison between the organic fraction of municipal solid waste from mechanical and source separation collection is shown on table 2.3.

The mechanically separated fraction from mixed waste of the MSW is characterised by a high content of dry solids, due to the inert fraction in the waste, which is incompletely separable with this approach and can also be mixed with size reduced organic matter (Table 2.3 - 2.4). This content of inert material can interfere in the equipment and also be present at the final product, reducing its value. If the inert content is removed prior digestion, it could contain size reduced organic matter that is going to be disposed of without treatment.

Table 2.3 – Comparison between the characteristics of mechanically separated (MS) and household source separated (SS) fraction of municipal solid waste (Barth, 2006; Favoino, 2005).

Composition (%)	MS	SS
Organic Matter	25.8	65.0
Paper	16.5	6.2
Plastic	4.6	2.3
Glass	4.3	0.7
Metals	1.3	0.2
Inert	3.5	0.5
Fabric	0.3	0.5
Particles (size<20mm)	43.8	24.7
Total	100	100
Dry Matter - DM (%)	46	35
Total Volatile Solid (%DM)	64	70

Table 2.4 – Mechanically selected organic fraction of municipal solid waste (Cecchi *et al*., 2002).

Composition (%)	Total Solid	Total Volatile Solid
Putrescible	59.0	78.0
Paper	4.6	7.1
Wood	1.1	2.2
Plastic	1.8	3.4
Inert	33.5	9.3
Total	100	100

2.6.1.1 Pre treatment - particle size reduction

The size of the particle can have a direct influence in the biological breakdown, overall temperature, moisture and mixing process. Several processes are available for particle size reduction; examples include trommels, shredders and ball mills (Archer *et al*., 2005). When the particle size reduction is applied in the up-front of the process it can lead to high level of contaminants such as heavy metals, plastics, glass, dust and bio-aerosols in the outputs - which could be dispersed throughout the organic fraction. The contaminant level varies according to the type of mechanical treatment used and the degree of aggressiveness applied so treatment influence the quality and the marketability of the output.

2.6.1.2 Post-refining

Post refining is applied in order to screen further undesirable materials (not biodegradable) for removal. Depending on the type of the application proposed for the output, the refining process is reduced or extended. Post-refining can also be used to change the output end-use application (pellets and bailing) (Archer *et al*., 2005). Examples of post-refining treatments are trommel, screening, vibrating screen, star screen, air separator.

2.6.2 Process outputs and applications

The anaerobic digestion process provides valuable outputs such as the biogas and the digestate (liquid, fibrous or solid), with several applications. Along with these products, a mechanically sorted stream with recyclables materials is also produced, with potential to

recover its value. This section will consider the application of the biogas and the solid digestate.

2.6.2.1 Biogas

The biogas is constituted mainly of methane and carbon dioxide, in an average composition of around 60% and 40%, respectively. The biogas can be used as energy and is the main source of revenue in the anaerobic treatment (Edelmann, 2003) as it is similar to natural gas, although with less calorific value, since the last is composed by different hydrocarbons (Wheeler and Rome, 2002).

The gas can be burned in a gas engine to generate electricity, which makes the anaerobic digestion plant self sufficient in energy and the surplus can be sold to the electricity grid. The generation of electricity can be applied more efficiently in a combined heat and power (CHP) system, where heat can be removed in the first instance to maintain the digester temperature (British Biogen, n.d.). A gas treatment is necessary prior use to eliminate contaminants compounds such as water vapour, ammonia and hydrogen sulfide, which is responsible for equipment corrosion (Edelmann, 2003). Another option is to upgrade the gas by removing the carbon dioxide for injection into the natural gas grid or for use in standard compressed natural gas (CNG) vehicles (Wheeler and Rome, 2002).

Connection to the national grid

The national grid is a network of power lines which allows distribution of electricity throughout the country. It can be connected to a single power source or electricity generating plant (it is usually linked with other plants to provide a more flexible/reliable

network). The electricity is usually transmitted at very high voltage, typically several hundred thousand volts, as this reduces losses and means that smaller cables can be used, reducing the overall cost of the network (REG, 2007).

The cost of grid connection is influenced by the voltage and proximity of the grid and whether there is a step down transformer already serving the area in question. Capital cost of the distribution system is very high varying from £20,000 - £60,000, for an 11 kV power line connection, but the maximum power generated and connected is less than 1 MW (additional costs for overhead lines are between £15,000 - £30,000 per km). For higher outputs of electricity (that would need 33 kV connections), connection equipment costs may range from £120,000 to £150,000 plus overhead line costs (£20,000 to £30,000 per km) (Lindegaard, 2007; REG, 2007; TSCTD, n.d.).

Biogas as biofuel

The use of biogas as biofuel is being implemented in European countries as an alternative to fossil fuels, in a process called up-rating, where the gas quality is improved to natural gas standards; the up-rating result is entitled renewable or sustainable natural gas (NGVA, 2007). The carbon dioxide emitted by burning renewable biogas has no added impact in the greenhouse effect since it is not a fossil fuel (Plombin, 2003). The only valuable fraction of the biogas for vehicle fuel is the methane, therefore, further treatment to eliminate carbon dioxide, water, sulphur and organo-halogens are necessary (Plombin, 2003).

Some countries also adopted the mixing of biogas with natural gas, for vehicle fuel. Natural gas is considered a clean fuel, in most countries in Europe, but it is not renewable; nevertheless it has less environmental impact than diesel (Table 2.5). Sweden is one of

the leading countries in the development of biogas fuelled vehicles with a fleet over 7.000 cars (plans to increase by 80.000 in 2010) as well as buses and train (EST, n.d.).

Table 2.5 – Comparison of gaseous emissions from heavy vehicles (Trafikkontoret, 2000)

Fuel	CO	HC*	NOx (g/km)	CO_2	Particles
Diesel	0.2	0.4	9.7	1053	0.1
Natural gas	0.4	0.6	1.1	524	<0.1
Biogas	0.1	0.4	5.4	223	<0.1

* hydrocarbons

Carbon dioxide application

Carbon dioxide is part of the biogas composition but since it does not have calorific value, this fraction is removed from the biogas and is released to atmosphere. Although CO_2 from renewable sources will not contribute to the increase of the greenhouse effect, it is important to develop market opportunities for it capture and utilisation as it will also allow other gas producers (fossil fuel-fired plants) to benefit from it. Several applications are being developed and some of then could be relevant for integrated waste management strategies:

- **Enhanced oil recovery:** is the dominant area for CO_2 utilisation. The gas is injected into an operational oil field, where it mixes with the crude oil, causing it to swell and become less viscous. This helps maintain reservoir pressures and oil production

rates. A considerable amount of the CO_2 injected remains in the formation over very long periods of time (IEA, 2003).

- **Food processing application:** high-purity CO_2 can be applied in the food and drink industries such as food processing duties, freezing/chilling purposes, beverage production. Such plants are already operating in the United States, Malaysia, Japan and Brazil (IEA, 2003).

- **Fertiliser manufacturer:** in some countries in Asia, CO_2 is captured (with amine scrubbers) and used in the production of urea (IEA, 2003).

- **Algae growth:** in Norway and Hawaii, the gas is used to growth algae for use as fish nutrients (IEA, 2003).

- **Enhanced plant growth:** elevated levels of CO_2 above natural (~250-330 ppm) can enhance the growth of plants and vegetables. Increase in yield up to 40% has been achieved in greenhouses with levels of 550 ppm (IEA, 2003).

For the future, emphasis will need to remain focused on ways to increase the range of possible application; improve the efficiency of those currently being used and confirm its technical and economic viability. The likelihood of such applications in the MBT context is unclear.

2.6.2.2 Solid digestate

The solid digestate from segregated waste collection is similar to aerobic compost and can be used as nutrient-rich soil conditioner spread on land for soil improvement and landscape restoration. This activity is preferable than traditional land spreading because it requires less equipment (Salminen and Rintala, 2002). The digestion treatment does not

reduce NPK (nitrogen, phosphorus, potassium) content, making the digestate valuable as a fertilizer (Salminen and Rintala, 2002; Wheeler, 2001). The fibre from AD can also be used as an alternative to peat, although it is not nutrient-free as peat (British Biogen, n.d.).

A possible strategy for increasing the value of the fibre is to further compost it aerobically after processing, enabling the product stabilisation, to produce a potting compost/growing medium. This process can be speeded up through careful management and control such as adding heat or insulating the composting bins (British Biogen, n.d.).

Solid residues coming from mixed waste collection might be referred as stabilised biowaste, to avoid confusion with compost from source separated materials. The possibilities of using the residue as compost into agricultural, horticultural or domestic applications in the UK are limited due to regulatory issues regarding the nature of the end product. It may also find barriers in the consumers as some contaminants (e.g.: plastics, glass, and metals) can be found in the final product. There are, however, different options for such material to be applied without compromising the area. Some options with relevance to the UK are as follow:

- **Land use to grow energy crops** is a limited application, since farmers could decide, in the future, to change back to use the land for food crops, which could compromise the quality of the land (Archer *et al.*, 2005).

- **Forestry** application is a possible application, as stabilised biowaste could be used as part of the growing medium, improving soil organic matter, water holding and nutrient supplying. It is likely that the biowaste producers are not going to generate income from its application (Archer *et al.*, 2005).

- **Verges and amenity lands** are a limited application in public area such as sport field and parks due to the possibility of contamination. There is a possibility to be

used on verge of trunk roads as long as the UK Environment Agency and the Scottish Environment Protection Agency do not consider such application as disposal (Archer *et al.*, 2005).

- **Landscaping and embankments** around road construction and others civil engineering projects are likely to be used (except for schools and housing), however, it is probable that constructions contractors would be paid to accept the material (Archer *et al.*, 2005).

- **Brownfield land redevelopment** is an attractive option, as the UK has thousands of sites classified in this category. Brownfield sites were previously contaminated by industrial use, and as the need for new developments is increasing, it will help to reduce pressure on greenfields for such purpose (Environment Agency, 2005).

- **Restoration for arid land** recovering the soil conditions, improving its quality and moisture retention, is a likely application, but with limited significance in the UK (Archer *et al.*, 2005).

- **Landfill disposal or daily cover** is the most likely application, as the material is going to have its biodegradability reduced, complying with Landfill Directive; however, landfill disposal can b considered as unsustainable and the landfill gate fees are increasing (Archer *et al*., 2005). Another option is to be used as a support medium for CH_4 oxidation, to mitigate greenhouse gas emissions (Einola *et al.*, 2008; Soyez and Plickert, 2002).

The application of stabilised biowaste from MBT process as a low grade material is limited, being mostly restricted to landfill disposal or as covering material, and is likely that it will not generate incomes for the producer; in contrast, it can imply in fees for its use. To

overcome this problem, the UK will need to develop quality standards for this material, which could provide understanding and assurance to the market.

2.7 Contaminants present in the feedstock

The presence of undesirable materials in the feedstock can interfere during the treatment process or in the value of the outputs. Contaminants from a mixed waste collection in a MBT plant can be originated from several sources and can be considered from heavy metals to aerosols. Depending on the aggressiveness of the mechanical technology applied, the levels of contaminants can increase significantly.

As previously discussed, solid contaminants presented in the feedstock are mainly plastics, scrap metals (ferrous and non-ferrous), glass and ceramics. During the mechanical treatment, a percentage of this material is removed as recyclables, therefore recovering its value. However, small particle size contaminants are not easily removed and depending on the biological treatment applied, these materials can be a problem. As an example, if anaerobic digestion is used, the presence of contaminants can block the pipelines or accumulate at the bottom of the reactor, leading to possible equipment damage.

2.7.1 Potential sources of air, water and land contamination

Emissions to air such as gases and bio-aerosols can arise from different parts of the process: when the biogas is combusted in a gas engine, exhaust gases containing pollutants are emitted into the atmosphere. During the mechanical treatment, fine particulates, bio-aerosols, odour (fugitive emissions) can also be released. By maintaining

a negative pressure within the plant, these emissions can be controlled, sending it to biofilters (Archer *et al.*, 2005). Heavy metals are unlikely to be a pollutant due to relatively low temperatures in the AD, as they would not be volatilised (Archer *et al.*, 2005).

The anaerobic digestion process can produce a liquid effluent (leachate) that can contain high levels of chemical oxygen demand (COD); a liquid residue will also be produced from the compost-like material, if the waste requires a post stabilisation (Archer *et al.*, 2005; Fricke *et al*. 2005).

Depending on the aggressiveness of the mechanical process, the digestate may contain size reduced contaminants (e.g.: glass, low grade plastics) as they can not be removed from the final product (Archer *et al.*, 2005). Heavy metals are also common contaminants in unsorted municipal solid waste, and can be found from several sources (Table 2.6); as they are not biodegradable they can accumulate to potentially toxic concentrations (Sterritt and Lester, 1980).

Pathogens can also be found at the end product and its presence or absence is due to the type of treatment used to reduce or eliminate these microorganisms. Treatment such as high temperatures is the most common option, occurring at the beginning or at the end of the biological treatment, avoiding the re-growth of microorganisms. Although pathogen reduction in anaerobic digestion is significant for bacteria, it is not satisfactory for viruses (Demuynck *et al.*, 1984), pathogenic spore-forming bacteria (such as *Clostridium botulinum, C. chauvoie and Bacillus anthracis*) and prions - proteinaceous infectious particle - responsible for transmissible spongiform encephalopathies (TSE) such as bovine spongiform encephalopathy (BSE) and scrapie (Sahlstrom, 2003).

Table 2.6 – Possible sources of trace heavy metals in materials found in MSW (Vassilev *et al.*, 1999).

Trace element	Potential Source
Mercury (Hg)	Batteries, plastics (PVC), fungicides, medicines, lamps, herbicides, pigments, paints, electronics, fluorescent tubes, alloys, galvanized items, fish remains
Cadmium (Cd)	Plastics stabilisers, papers, paint, pigments, batteries, printing inks, galvanised items, alloys, solders, surface metal coatings, textiles, semiconductors, glazed ceramics
Thallium (Tl)	Electronics, semiconductors, optics, certain types of glass, fuels, lamps, alloys, plant materials, biological tissues
Arsenic (As)	Clay materials, paints, medicines, pesticides, electronics, semiconductors, cosmetics, certain glass types, alloys, lamps, leather, orchard leaves
Antimony (Sb)	Plastics, alloys, electronics, semiconductors, batteries, rubber, pigments, textiles, cables, surface metal coatings, certain types of glass, medicines
Copper (Cu)	Alloys, steels, electronics, papers, printing materials, paints, plastics, galvanized items, building materials, fungicides, plant matter, chicken plasma
Cobalt (Co)	Alloys, steels, inks, magnets, fuels, pigments, ceramics, certain types of glass, fertilisers
Chromium (Cr)	Cardboards, papers, certain types of glass, paints, pigments, leather, alloys, steels, electronics, surface metal coatings, galvanized items, fireproofing, plastics
Nickel (Ni)	Alloys, steels, batteries, plastics, pigments, certain types of glass, coins, electronics, surface metal coatings, magnets, vegetable oils
Molybdenum (Mo)	Alloys, steels, batteries, electronics, lamps, papers
Zinc (Zn)	Alloys, printing inks, papers, rubber, plastic, batteries, surface metal coatings, galvanized items, pigments, semiconductors, pesticides, medicines, food remains
Tin (Sn)	Plastics, stabilisers, tins, solders, surface metal coatings, galvanized items, pigments, semiconductors, pesticides, medicines, food remains
Lead (Pb)	Plastics, pipes, paints, pigments, alloys, papers, cardboards, rubber, batteries, printing inks, glazed ceramics, electronics, cables, solders, surface metal coatings, galvanized items, certain types of glass, fuels, food remains, blood
Manganese (Mn)	Steels, alloys, batteries, certain types of glass, resins, pigments, galvanized items, fuels, textiles, pesticides, fungicides, fertilisers, fatty acids
Vanadium (V)	Steels, alloys, electronics, textiles, varnishes, rubber, ceramics, certain types of glass, medicines

2.8 Legislation

This section reviews different policy instruments that will affect and drive the implementation of MBT and anaerobic digestion treatment in the UK. Also it focuses in the use of the outputs – digestate and biogas.

2.8.1 Waste Framework Directive

The Council Directive on Waste (EC 75/442/EEC, 2003), controls the disposal and recovery of waste across the European Community. It gives priority to waste prevention and encourage reuse and recovery practices. According to the Directive waste recovery operation such as spreading waste on land, which results in benefit to land, is classified as land treatment activity. When there is no benefit to agriculture or ecological improvement, the activity is considered as waste disposal (Alker, 2004). Therefore, the application of digestate residues from MBT on land has the potential to be considered as a recovery operation, but with exemptions.

2.8.2 Landfill Directive

The EU Landfill Directive (1999/31/EC, 1999) imposes restrictions on the amount of biological municipal waste to landfill, aiming to prevent or reduce its negative impact in the environment (Archer *et al.*, 2005; DEFRA, 2006). The Directive was included into national legislation by the Landfill Regulations (England and Wales, SI 2002/1559; Scotland, SSI 2003/235; Northern Ireland, SR 2003/496).

The Directive sets challenging targets to reduce the amount of BMW sent to landfill. As consequence, England will have to divert 25% of the waste produced by 2010, 50% by 2013 and 65% by 2020, when compared to 1999 levels, as reference year. The Directive also promotes the creation of products from the waste diverted from landfill (Alker, 2004; Hawkins and Shaw, 2004).

The increasing stringent Landfill Directive targets can strongly benefit the implementation of MBT and anaerobic digestion, as the main objective of these processes are the diversion and treatment of biodegradable municipal waste, and consequently its volume, as well as, for MBT, the recovery of value from recyclable materials.

2.8.3 Landfill Allowances and Trading Scheme Regulation

Landfill Allowances and Trading Scheme (LATS or LAS in Scotland) was introduced in the Waste and Emissions Trading Act (2003) and launched in the UK in 2004 (England, SI 2004/3212; Scotland, SSI 2005/157; Wales, WSI 2004/1490; Northern Ireland, SR 2004/416) and is a flexible economic instrument for local authorities (LA). Each local authority was allocated with annual allowances, based on waste arising and waste sent to landfill, recycling, composting or recovery, in 2001/02; each allowance consents a LA to landfill one tonne of BMW. Allowances not used in the year can be banked or sold, generating extra income; authorities in need for extra disposal, will have to buy allowances from neighbours LA. The breach in the allowance target will incur fixed penalty of £150/tonne (Archer *et al.*, 2005).

The implementation of LATS should assist authorities to find cost effective techniques to meet their targets as well as benefiting authorities with diversion system already in practice, providing an alternative income. The LATS scheme will therefore promote the

implementation of waste treatment process able to reduce the biodegradability of the waste, such as MBT and anaerobic digestion.

2.8.4 Animal by Products Regulations (ABPR)

The Animal By-Product Regulation (EC 1774/2002, 2002) introduced in the UK in 2003 (SI 2003/1482; SSI 2003/411; WSI 2003/2756; SR 2003/495) came into force to guarantee collection, transport, storage, handling, processing and use or disposal that do not put in risk human or environmental health (Kirchmayr *et al.*, 2003). The regulation defined animal by-product as “any part of an animal carcass, or any material of animal origin, not intended for human consumption”. The regulation establishes 3 different categories of animal by-products:

Category 1: include materials with the highest risk for public health, animals or environment (risk of spread TSE diseases) and must not be processed in a biogas plant (Kirchmayr *et al.*, 2003). The materials are catering waste from international means of transport, animals and part of animals suspected of being infected or killed in context of TSE (EC 1774/2002, 2002). These materials can be incinerated or disposed of in landfills (Nordberg, 2004).

Category 2: include all animal by-products which can be allocated neither to category 1 nor to category 3 (Kirchmayr *et al.*, 2003). Examples of this material are digestive tract content, manure, milk not fit for human consumption, fallen animals, and solid materials from slaughterhouse (EC 1774/2002, 2002). These materials can be digested but has to pass a heat treatment unit at 133˚C for at least 20 minutes (or equal treatment) before pasteurisation (EC 1774/2002, 2002; Nordberg, 2004).

Category 3: include those animal by-products which would be fit for human consumption, but are - for commercial reasons - not intended for human consumption, such as catering waste, meat-containing waste from foodstuff industry (Kirchmayr *et al.*, 2003). Materials can be fed to an approved biogas plant after sterilisation with steam pressure. Approval conditions are found in the Article 15 of the ABP-Regulation (Nordberg, 2004).

As the source of feedstock for MBT is mainly unsorted waste, the possibility of output contamination is high, being in disfavour for public use. For landfill application, it is possible that other technologies could be incorporated in the MBT design.

2.8.5 Renewable Obligation Order

The Renewable Obligation Order was introduced in 2002 (England and Wales, SI 2002/914; Scotland, SSI 2005/185, Northern Ireland, SR 2006/56) and places an obligation on all licensed electricity suppliers to produce or buy from outside generators, an annually increasing percentage of their total sales from eligible renewable sources. The order aims to create a substantial demand for renewables (Martin, 2003), as suppliers must achieve 5.5% for 2005-06 rising to 10.4% by 2010-11 and 15.4% by 2015-16 (SSEFRA, 2006). The Obligation will remain in place until 2027, to guarantee a stable and long term market.

Renewable Obligation Certificate (ROC) is awarded to accredited generators of eligible renewable electricity produced within the UK per each ‘megawatt per hour’ provided; additional revenue is offered to waste treatment facilities able to generate renewable electricity. Companies also have the option of “buying out” their obligation. The price was initially set at £30/MWh for 2003. In 2004 it was sold in auctions by the record average price of £52/MWh (NFPA, 2004) while in the first semester of 2006, the auction average

price was £40/MWh (NFPA, 2006). The price achieved in the auction on October 2007 was £49/MWh (NFPA, 2006).

The Renewable Obligation supports energy from waste technologies, so MBT that uses anaerobic digestion process will benefit, since the electricity surplus can be sold generating extra revenue.

2.8.6 Pollution Prevention and Control (PPC)

The Pollution Prevention Control (England and Wales, SI 2000/1973; Scotland, SSI 2000/323) is a regime to control pollution from waste activities, introducing the concept of best available technique (BAT) to environmental regulations. It implements the European Directive on Integrated Pollution Prevention Control (EC/96/61) and aims to prevent pollution to air, land and water and balance the costs to the operator against benefice to the environment.

The BAT reference document, 'Waste Treatment Industries', was elaborated in 2005 making suggestions and recommendations for anaerobic digestion and MBT. The IPPC Directive will be used to regulate these two processes, which will be advantageous, especially for the new developments, since they will have their design compliant, in comparison to older plants.

2.8.7 Publicly Available Specification 100 (PAS100)

The British Standard Institute PAS 100 is a compost specification that requires the quality assessment of the outputs for the levels of pathogens, heavy metals, phytotoxins, carbon:nitrogen ratio, prior the use on land. At the present, outputs from mixed waste

collection (as MBT) cannot be certified under PAS100, but it can be used as benchmark. Being a voluntary standard, PAS 100 does not prohibit the marketing of MBT outputs; however, such applications will be constrained significantly (Archer *et al.*, 2005).

2.8.8 Biofuels Directive

The Biofuels Directive (2003/30/EC) intends to replace the use of petrol and diesel for biofuels and other renewable fuels for transport. In doing so, it contributes to meeting climate change objectives, promote renewable energy sources and provide an environmentally friendly secure supply. Member States were required to set national indicative targets for 2005 and 2010 for the minimum proportion of renewable fuels, against reference values of 2% and 5.75%, respectively, by energy content (DFT, 2004).

According to Deurwaarder (2005), the total sales of biofuels in the UK were around 19.5 million litres in 2003, corresponding to approximately 0.04% of the total road transport fuels. It was estimated that with the policy measures and additional incentives, the UK biofuels sales would be around 144 million litres in 2005, corresponding to 0.3% of the total fuel produced. The Directive, however, could adversely promote the deforestation for energy crops, decrease food crops cultivation, and increase levels of carbon dioxide that will be in disagreement with its primary objective (Deurwaarder, 2005).

In Sweden and Switzerland, biogas is already being used as an alternative to fuel (Cecchi and Battistoni, 2002). In the UK, although great support is being dedicated to alternative fuels such as hydrogen, biodiesel, biomass, natural gas and LPG - liquefied petroleum gas (DTI, 2003) it is expected that biogas can also be positively affected by the Directive.

2.8.9 Packaging and Packaging Waste Directive

The Packaging and Packaging Waste Directive (94/62/EC) defined targets for the recycling/recovery of materials as follow: 60% recovery of glass; 50% recovery of metal and 22.5% recovery of plastic by 2008. Although the percentage of recyclable recovered in the MBT process is low when compared to source separated collection, MBT can still benefit from the Directive, contributing to achieve the targets.

2.8.10 Waste Strategy for England 2007

The Waste Strategy for England, together with Planning Policy Statement 10 *Planning for Sustainable Waste Management* (PPS10) is part of the implementation for England of the requirements within the Framework Directive on Waste, and associated Directives (Hazardous Waste and Packaging Waste Directive), to produce waste management plans. It is also a strategy for dealing with waste diverted from landfill, as required by the Landfill Directive.

The Waste Strategy 2007 emphasises its support to the implementation of anaerobic digestion as energy recovery process through new technologies programmes. It also mentions the establishment of an Anaerobic Digestion Policy Network, by DEFRA, to take forward work on the technology and maximise the synergies between the different markets for it. Regarding MBT, the Strategy mentions that decisions made by local authorities and stakeholders will be crucial to the development of this treatment. There are also new higher national targets for recycling and composting of household waste – 40% by 2010, 45% by 2015 and 50% by 2020 – and recovery of municipal waste – 53% by 2010, 67% by 2015 and 75% by 2020 – which will require the implementation of broad technologies to delivery it.

2.8.11 Biowaste Directive

The EU Biowaste Directive was planned to be issued by the end of 2004 with the aim to control potential contamination and to encourage the use of certified compost, by setting quality standards, ensuring long-term safe applications. The Directive would also emphasise the role of anaerobic digestion and mechanical-biological treatment to reduce the quantity of biodegradable waste being lanfilled (ASSURRE, 2004; ESA, 2004). However, the Directive was abandoned and the content to be incorporated by further Directives.

In 2008, the European Parliament Agriculture Committee proposed the development of a possibly joint EU directive on biogas and biowaste, targeting between others, the national and regional planning measures to reduce barriers as well as incentive to invest in biogas plants (ESA, 2008).

CHAPTER 3

MATERIALS & METHODS

3.1 Physico-chemical characteristics of the feedstock

The waste samples were collected from two large-scale MBT plants in the UK treating mixed municipal solid waste. The option for each plant was considered according to individual design and technical process applied so as to obtain waste samples after the mechanical treatment stage. Technical information was provided by the MBT report from Juniper (Archer *et al.*, 2005) and further consulting with site operators.

Due to limited number of plants available in the UK with the desired characteristics at the time of collection, just one of the plants chosen had anaerobic digestion as biological stage; the other plant applied aerobic composting, therefore, the waste supplied presented higher particle sizes than the one using anaerobic digestion. The waste samples were collected on each site after particle size reduction and removal of recyclables materials such as plastics and metals and prior biological treatment. A description of the mechanical process used by each company is presented on appendix A.

As it is not on the scope of this project to compare or analyse the performance of individual waste treatment plant, the selected waste streams from MBT plants with anaerobic digestion (site 1) and with aerobic composting (site 2) will be indentified hereafter as MBT-1 and MBT-2, respectively.

In order to further remove materials recalcitrant to biodegradation, such as plastic, glass and ferrous and non-ferrous metals and homogenise the samples (Fig 3.1), the wastes were separated in a 0.6 cm^2 sieve for MBT-1 waste and in a 2.5 and 0.6 cm^2 sieves, for

the MBT-2 waste. The test sample (i.e.: sample sent to the laboratory that is further prepared for testing or analysis) was prepared according to British Standard (BS EN 15002:2006). The sample was manually pre-homogenised using a heavy-duty scoop; remaining contaminants were manually removed using forceps; a sub-sampling technique (coning and quartering) was used to divide the waste into different representative portions. The final portion was collected and mechanically homogenised with a manual blender.

The eluate (i.e.: solution remaining after the laboratory leaching procedure of a solid material in contact with a leachant) was prepared according to British Standard (BS EN 12457-4), by mixing the test sample previously prepared with de-ionised water at 10 L/kg (dry matter) dilution. The solution was placed in a roller table for 24 hours and further centrifuged at relative centrifugal force (RCF) of 3310 g for 12 minutes; the liquid fraction resulting was filtered with vacuum process.

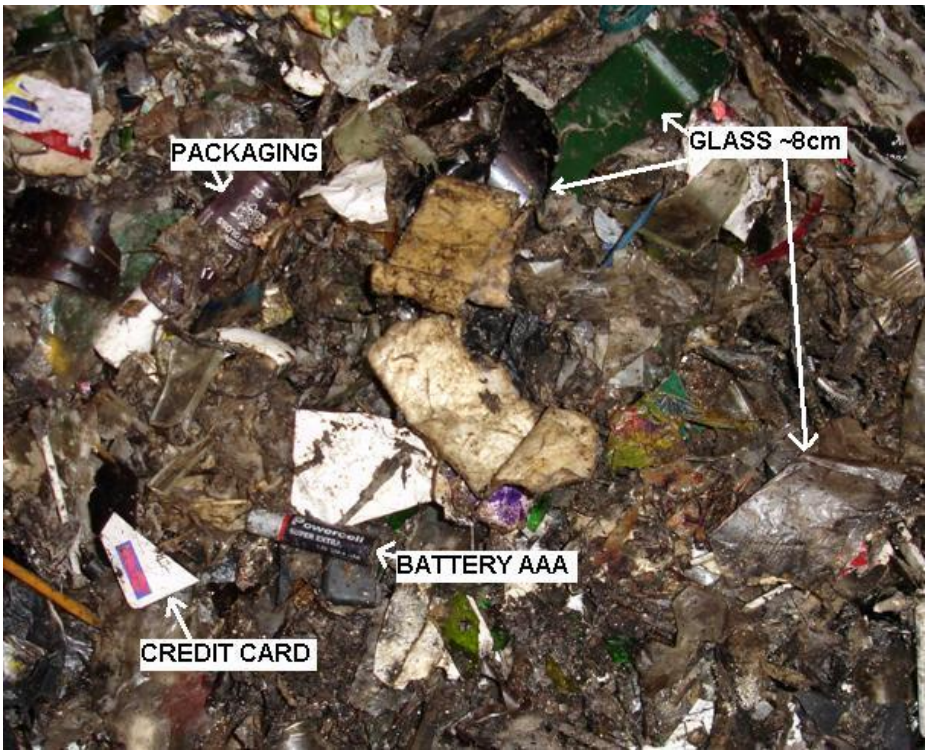

Figure 3.1 – Example of contaminants removed at the laboratory.

Gravimetric analysis (in triplicates) was performed in the test sample to determine dry matter and moisture content, by placing the sample in the oven and drying overnight at 105 ℃, and to determine volatile solids and fixed solids, by placing the previously dried sample in a muffle furnace, at 550 ℃ for one hour (loss on ignition).

Chemical analysis (in triplicates) was conducted in the previously prepared eluate to analyse chemical oxygen demand (COD); total nitrogen; volatile acids; pH and potential redox. The parameters total carbon, total organic carbon and total nitrogen (all in %) were analysed using the test sample, in an elemental analyser (*Vario EL Elementar Analysensysteme GmbH, Germany*), through catalytic tube combustion in oxygenated CO_2 atmosphere with high temperatures.

Chemical oxygen demand (COD) analysis was carried out using the test kit *COD* (*LCK014 Hach Lange* - 1000-10,000 mg/L) following the recommendation of the ISO 8466-1 (1990) method. The total nitrogen was analysed following the instructions of the test kit *Nitrogen (total) Cell Test* (*Spectroquant* 1.14763.0001). The pH and conductivity were measured in a *Jenway 3540 pH and Conductivity Meter*; the redox was measured in a *Jenway Ion Meter 3345*. Alkalinity was measured by titration with end point at pH 4.5, using HCl (0.02 M) as titrant.

3.2 Anaerobic digestion set up

3.2.1 Semi-continuous loading system

Six 5-litres (working volume) stainless steel (grade 316) continuous stirred tank reactor (CSTR) chemostats, were set up in semi-continuous regime at mesophilic temperatures (35 ˚C) in a cabin-size laboratory, at Summerleaze AnDigestion (Waterbeach, Cambridgeshire). Each reactor was equipped with mixing paddle device, constantly stirring

at 60 rotations per minute (rpm). An individual temperature probe was connected to a control panel in conjunction with an electric blanket (*Holroy Components*), to control and ensure adequate temperature. The biogas production from each reactor was measured using a gas flow-meter, consisted of an acrylic container with an internal rotating chamber divided in half (Figures 3.2-3.3), filled with acidified water (pH ≤ 3) to avoid CO_2 dilution. The gas was released from the digesters and when full, the chamber rotated 90˚, releasing the gas. At each rotation, the equipment emitted an electrical signal that was computed in a digital counter. Each rotating chamber had its internal volume pre determined so as to relate the numbers of tipping with the amount of gas produced.

From the flow meter, the biogas was collected in 13 litres Tedlar bags; the biogas content - CH_4, CO_2, O_2 (%) H_2S and CO, in parts per million (ppm) were measured (in triplicates) on a regular basis using a calibrated *GA 2000* infrared analyser *(Geotechnical Instruments, UK)*. The analyser was the same used by Summerleaze to monitor the biogas produced at Donarbon landfill site, therefore complying with regulation. For the measurement, the bags were closed and disconnected from the flow meter and connected to the analyser. The data were recorded after enough gas was constantly pumped (minimum 2 minutes) so as to have a stable reading. After reading, the bags were connected to an air pump in order to empty out the gas content and re-connected to the flow meter. Daily control involved the measurement of temperature and pH in the digestate, biogas production and composition. At the end of each trial, the success of the digestion was determined by the quantity and quality of biogas produced and level of waste reduced.

Prior the digestion trial a start up process was elaborated in order to acclimatise the microorganisms to the feed and stabilise the system, allowing an optimum bacteria content. It also helped to verify possible problems with the set up and make necessary adjustments, since it was the first time it was being used. During the start up, temperature,

pH and biogas quality were constantly monitored and controlled. A successful start up was determined when a stable biogas production ($CH_4 \geq 50\%$) was achieved.

Approximately 250 mL of digestate from each reactor were daily removed to measure temperature and pH (*WTW - Multi 340i*). Waste material MBT-1 was used as fed, in a loading system of 3 gVS/L; the waste was weigh, mixed with water and blended for approximately 5 seconds, in order to homogenise and facilitate the feeding process. The hydraulic retention time (HRT) was set initially at 30 days (HRT = reactor volume/flow rate). To maintain and constant volume, the same volume added was discarded from the digestate, and the remaining was filtered and added to the feed so as the final volume added was waste + water + digestate = 167 mL. The filtered portion was weigh and placed to dry overnight in the oven at 105 ℃, determining its moisture content; the dried sample was further placed in a muffle furnace, at 550 ℃ for one hour to determine volatile solids and fixed solids (loss on ignition). The data related to the quantity of waste in and out of the system enabled the mass balance analysis.

Three anaerobic digestion trials were proposed in order to investigate different parameters that could influence in the anaerobic digestion performance: 1) loading rate at 3 gVS/L and 30 days hydraulic retention time; 2) loading rate at 3 gVS/L and 25 days hydraulic retention time; 3) loading rate at 4.5 gVS/L, 20 days hydraulic retention time and two different waste streams. Six reactors were operating at the same condition during trials 1 and 2 while three reactors were operating at the same condition during trial 3. The reactors were filled-up with digestate from anaerobic digester treating municipal waste from Summerleaze AnDigestion (Holsworthy, North Devon). Heavy metal concentration and microbial community profile were determined during the third trial, when two different waste streams were used as feedstock (Table 3.1).

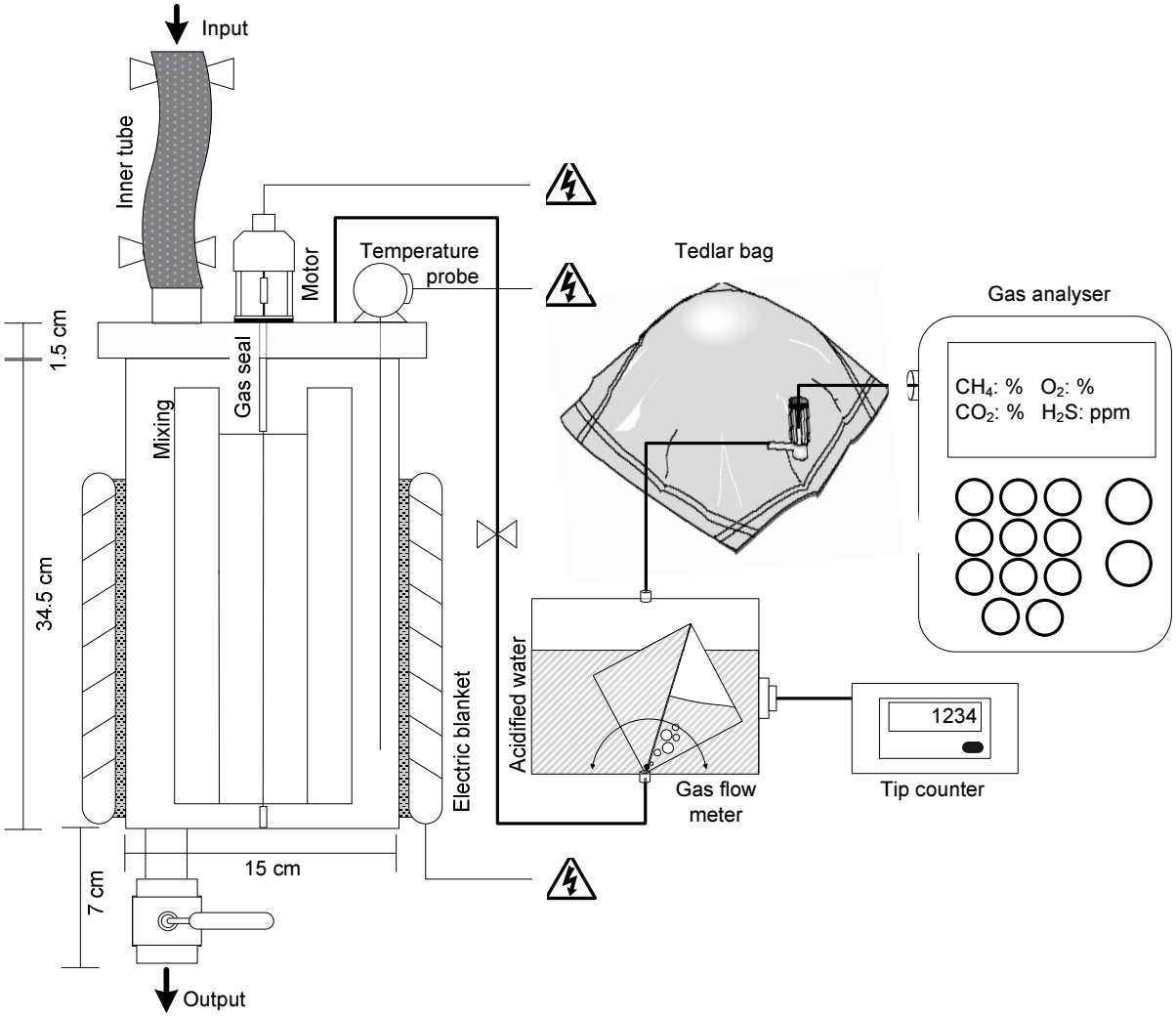

Figure 3.2 - Anaerobic digestion set up – continuous.

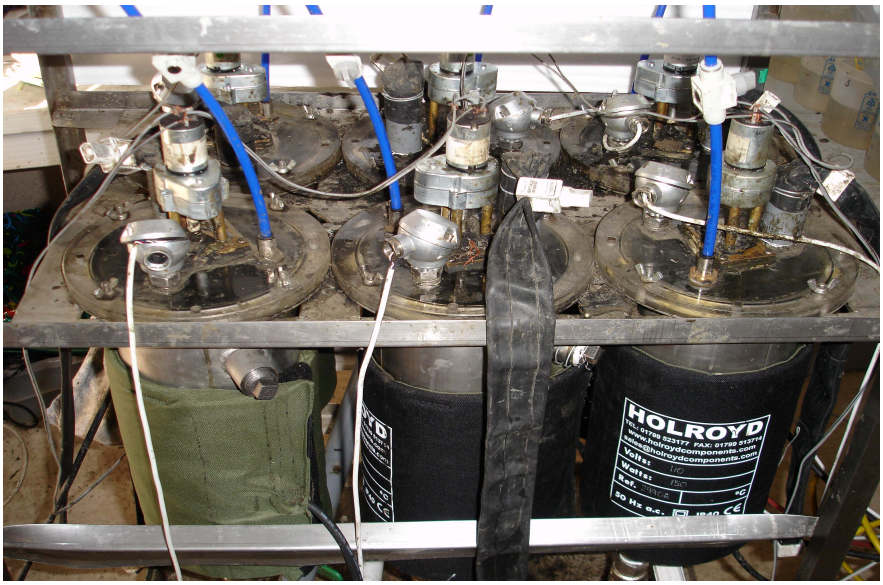

Figure 3.3 - Anaerobic digestion set up – continuous.

Table 3.1 – Summary of the activities during the trials.

Trials	Start up	1	2	3
Duration (days)	25	32	36	46
Loading rate	2.5 gVS/L	2.5 gVS/L	3 gVS/L	4.5 gVS/L
HRT*	30	30	25	20
Feedstock used	1	1	1	2
Type of analysis	biogas, temperature, pH	biogas, temperature, pH	biogas, temperature, pH	biogas, temperature, pH, heavy metal, microbiology

* HRT: hydraulic retention time

3.2.2 Batch loading system

The batch trials consisted of eight 1-litre glass bottle reactors (working volume: 0.5 litre) incubated in a water bath (*Fischer Scientific*), at mesophilic temperatures (35 ˚C), with an initial loading of 12 gVS/L. The size of the reactor was similar to the one used in the Environment Agency guidance for MBT (2005) and also comparable to those reported in the study carried out by Zhang *et al.* (2007) and Chen *et al.* (2004). Each waste stream was digested in triplicate with a blank control. All reactors were filled with 300 mL of acclimatised seed (digestate from Summerleaze biogas plant - Holsworthy) and filled up to 500 mL (working volume) with water; nitrogen gas was purged in each reactor to assure anaerobic conditions. The bottles were manually mixed on a regular basis allowing the biogas to be released and be captured in water displacement set up, in Mariotte flasks containing acidified water (pH ≤ 3) to avoid dissolving CO_2 in the water (Figures 3.4-3.5).

After mixing, the gas volume produced was recorded, collected with an adapted syringe and purged in the infrared gas analyser (*GA2000 Geotechnical Instruments, UK*). Due to instrument limitation, a minimum of 60 mL had to be purged to obtain a reliable reading, so the biogas was not collected until this minimum amount was produced. The production of biogas from the digesters was adjusted with the biogas produced from the blank, containing just seed, so as the final value corresponded to the gas produced from the waste solely. The experiment took place until insignificant or zero amount of gas was produced.

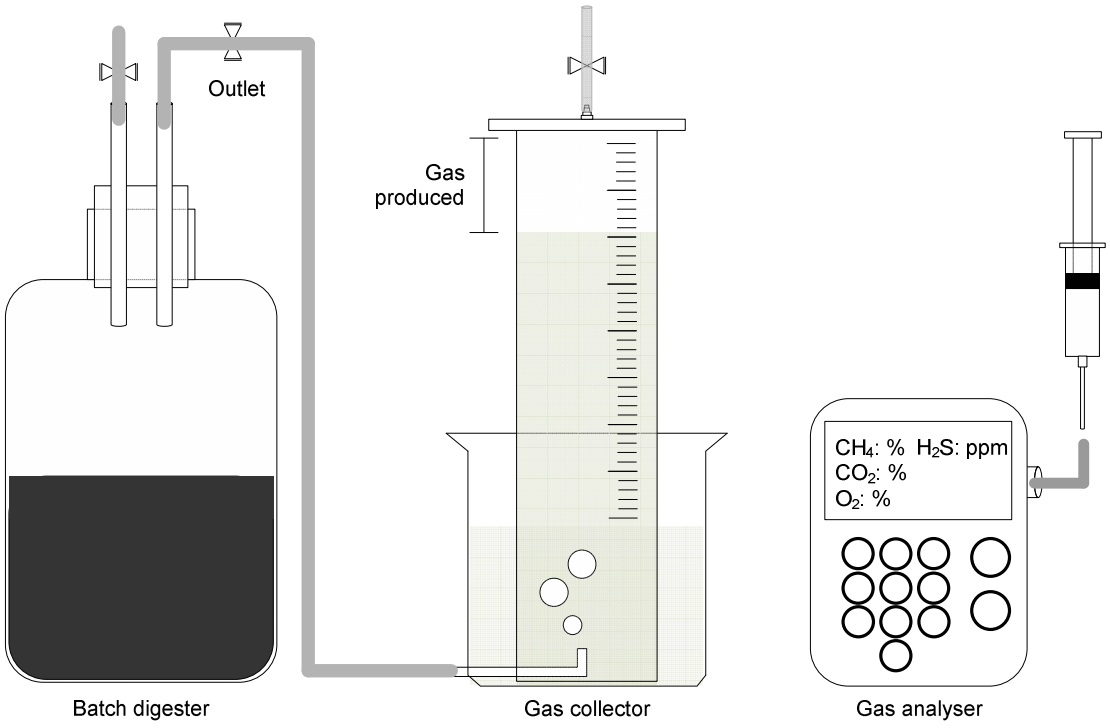

Figure 3.4 – Anaerobic digestion set up – batch.

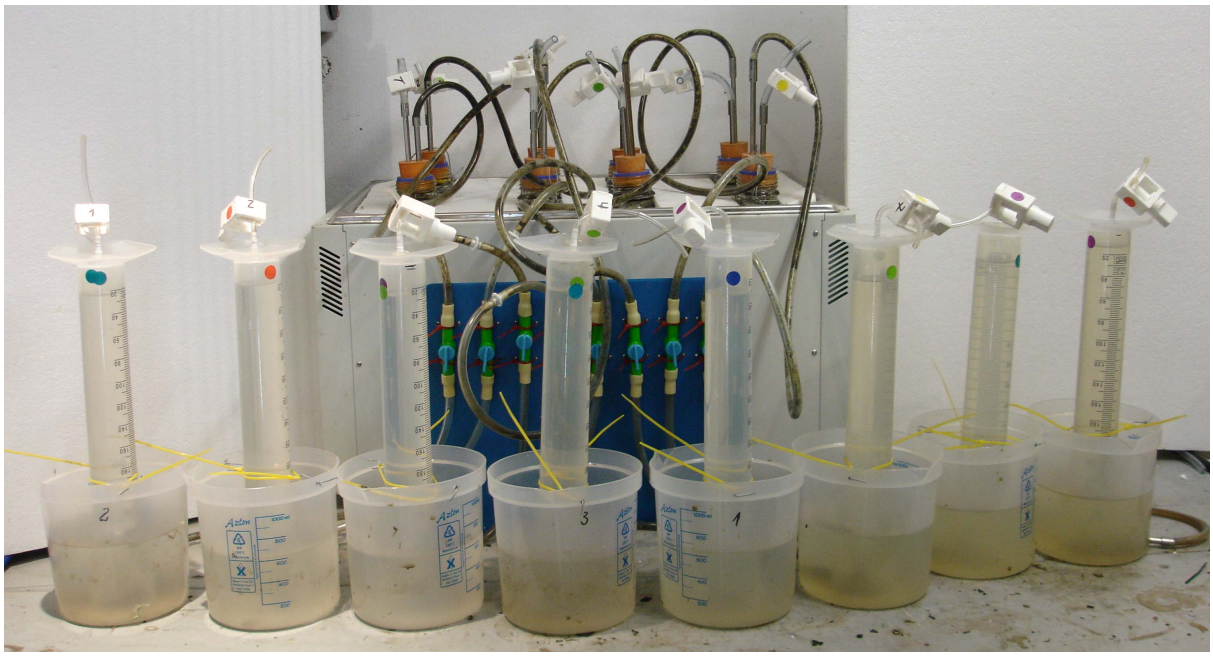

Figure 3.5 – Anaerobic digestion set up – batch.

3.3 Heavy metal concentration

Heavy metals cadmium (Cd); chromium (Cr); nickel (Ni); copper (Cu); lead (Pb) and zinc (Zn) were analysed (in triplicate) during the third experiment, when MBT-2 started to be used as feedstock. Digestate samples were collected at day 0, 15 and 30, and kept frozen (-20 ˚C) prior analysis; after defrosting, they were centrifuged at relative centrifugal force of 600 g for 5 minutes, and vacuum filtered. Prior analysis, samples were diluted (1:2) with ultra pure water. Heavy metals detection and calibration range used, in µg/L, were: 0-2 and 0-100 (Cd), 0-35 and 0-50 (Cr), 0-356 and 0-500 (Ni), 0-65 and 0-100 (Cu), 0-8 and 0-10 (Pb), 0-86 and 0-100 (Zn). The functionality of the ICP-MS was checked by following the manufacturer recommended daily optimisations and followed by a daily performance test. The heavy metals concentration was analysed with inductively coupled plasma mass spectrometry – ICP-MS (*Perkin Elmer Elan 9000*, *UK*), with sample flow rate of 1 mL/min. Mixed standards were prepared from commercially available single metal

standard solutions (*Fisher Scientific, UK*) and diluted in ultrapure water (*Elga, UK*). An internal standard of rhodium was added to both standards and sample solutions. The isotopes of each element used were cadmium 114; chromium 50; nickel 58; copper 65; lead 208 and zinc 66.

3.4 Microbial community analysis

3.4.1 Phospholipids fatty acids analysis

Phospholipid fatty acids (PLFA), due to its high biological specificity, provide the description of a microbial community at a particular time, as well as the community shift (population dynamics) through different operational conditions (Narihiro and Sekiguchi, 2007; Silvey et al., 2000; White, 1983; Balkwill *et al.*, 1988; Tunlid and White, 1990). PLFA profiles were determined using a modification of the method described by Frostegard *et al.* (1991), based on the method described by Bligh and Dyer (1952) and White *et al.* (1979). PLFA were extracted from an equivalent to 10 mL digestate samples, stored frozen at -80 ℃ and freeze dried.

Sample was transferred to a glass tube with a Teflon-lined screw cap and added with 15 mL of Bligh and Dyer (B+D) solvent: 0.15 M citrate buffer, chloroform, methanol, in a 0.8:1:2 ratio, 0.005% w/v (50 mg/L) 2,6,di-tert butyl-4-methylphenol as anti-oxidant. Citrate Buffer: 0.15 M of citric acid dihydrated (31.5 g/L) and 0.15 M of trisodium citrate (44.1 g/L) in deionised water adjusted to pH 4 using diluted acetic acid. Samples were sonicated 30 min, shaken in a roller table 30 min, and centrifuged at relative centrifugal force (RCF) of 883 g (*Falcon 6/300 Refrigerated Centrifuge - MSE, UK*).

The organic layer was removed into a clean glass media bottle and separated into 2 phases by adding 4 mL of chloroform and 4 mL citrate buffer, followed by further centrifugation. The organic layer was dried under a stream of N_2 at 37 ºC. Polar lipids were separated from neutral and glycolipids using solid-phase extraction (3 mL/500 mg Si *Sep-pak Vac*^{TM} *- Waters Chromatography, UK*), by elution with 5 mL chloroform, followed by elution of the 12 mL of acetone. Polar lipids (including the phospholipids) were then eluted with 8 mL of methanol, and dried under N_2 stream at 37 ℃.

The resulting polar lipid fraction was then subjected to mild alkaline methanolysis according Dowling *et al.* (1886). Samples were reconstituted with toluene:methanol 1 mL (1:1) and methanolic potassium hydroxide (0.56 g KOH/50 mL) 1 mL (0.2 M), agitated and incubated at 37 ℃ for 30 minutes. Then acetic acid 0.25 mL (1 M), hexam:chloroform 5 mL (4:1 v/v) and deionised water 3 mL were added, followed by sonication 30 minutes and centrifugation. The remaining fraction was washed with sodium hydroxide 3 mL (0.3 M), filtered through sodium sulfate, evaporated under a stream of N_2 at room temperature, reconstituted with hexane 200 µL, transferred into a GC vial.

FAMEs were analyzed on a Gas Chromatograph (Agilent Technologies, UK, Model 6890N) fitted with a HP-5 (Agilent Technologies) capillary columm of length 30 m, internal diameter 0.32 and film thickness 0.25 µm, 5% phenylmethyl-siloxane. Carrier gas used was helium at a velocity of 1 mL/min. Samples of 1 µL were injected with an autosampler in splitless mode at 310 ℃. The oven temperature started at 50 ˚C for 1 minute, and increased to 160 ˚C at 25 ˚C/min, then increased to 240 ˚C at 2 ˚C /min, and further increased to 310 ˚C at 25 ˚C/min. FAMEs were detected using flame ionization detector at 320 ℃, and identified by comparing retention times. The chromatograms were used to quantify FAME by peak area. Each FAME in a chromatogram (area ≥ 15) was quantified on relative percent by weight basis (manufacturer recommendation) by dividing the peak area corresponding to the FAME by the total area of all the component FAME peaks.

The results were organized through principal component analysis to reduce the data set to lower dimensions; the analysis provided a correlation matrix with eigenvalues, where the factors to be examined were determined. The statistical analysis to compare community profile shifts during time and between the reactors was conducted using repeated measures of analysis of variance (ANOVA) from Statistica software (*StatSoft, 2007, version 8.0*). Cluster analysis based on presence/absence (binary matrix) of PLFA peaks was then carried out using the beta version of Primer 6 (Clarke and Warwick, 2001) by performing a hierarchical cluster analysis based on Bray-Curtis similarity matrix.

CHAPTER 4

RESULTS & DISCUSSION

4.1 Initial characterisation

General chemical and physical properties of the waste samples demonstrated similarities between both materials (Table 4.1). Levels of dry matter in both samples were comparable to those found in similar experiments with municipal solid waste (Blanco, 2008; Pahl, *et al.*, 2007, Favoino, 2005; Barth, 2006, Ponsá *et al.*, 2008); however, these values indicated the presence of high content of inert material in the waste, which could result in accumulation inside the reactors. The levels of volatile solids served as an estimative for the biogas potential of the waste, and are according to research conducted by (Pahl, *et al.*, 2007, Favoino, 2005; Barth, 2006, Ponsá *et al.*, 2008) indicating acceptable conditions for the digestion. The C:N ratio for both materials also indicate ideal conditions for the digestion, according to Monnet (2003). Presence of N (nitrate+nitrite) was also important as organic contaminants can be degraded under anaerobic conditions with a wide variety of terminal electron acceptors, such as nitrate (Burland and Edwards, 1999), chlorate (Weelink *et al*., 2007), iron (Lovley *et al.*, 1994), manganese (Villatoro-Monzón *et al.*, 2003), sulphate (Lovley *et al.*, 1995) and under methanogenic conditions (Grbić-Galić and Vogel, 1987) and even by pure cultures (Coates *et al.*, 2001; Kasai *et al*., 2006).

Table 4.1 – Initial waste characterisation

Waste Characterisation	MBT-1 (±sd)*	MBT-2 (±sd)
Moisture Content (%)	49 (0.4)	53 (1.7)
Total Solids (%)	52 (0.4)	47 (1.7)
Total Fixed Solids (%TS)	38 (1.7)	38 (4.6)
Total Volatile Solids (%TS)	63 (1.7)	62 (4.6)
Total Volatile Solids (%total weigh)	32 (0.7)	29 (1.1)
pH	7.4 (0.1)	5.1 (0.08)
Redox (mV)	245	272
Conductivity (µS)	3.1 (0.4)	5.9 (0.8)
COD (mg/L)	1668 (87)	11716 (817)
Total Carbon (%)	32 (1.0)	39 (0.1)
Total Organic Carbon (%)	29 (3.2)	37 (0.1)
Total Nitrogen (%)	1.3 (0.04)	1.6 (0.02)
C:N ratio	25	24
Alkalinity (mg $CaCO_3$/L)	1740 (110)	1175 (82)

*sd: standard deviation.

4.2 Anaerobic digestion

4.2.1 Semi-continuous loading system - chemostat reactors

4.2.1.1 Mechanical adjustment during reactors start-up

The start-up process helped to verify possible problems with the chemostat set-up and make necessary adjustments. The first adjustment was regarding logistic for the input of feed. The original set up consisted of a 50 cm vertical PVC tube with a valve in the middle; the length of the tube was high due to special impediment of the surrounding fittings on the top of the reactor. The upper part was fed to the top, with the valve off, and an adapted plunger was used to seal it and push the content inside the reactor, when the valve was turned on. Levels of oxygen ranging from 1 to 2% were found inside the reactor. The problem was solved by changing the PVC apparatus to a bicycle inner tube with plastic screw clamps at the two extremes. This way, the feed was placed inside the tube with the bottom valve shut; the air was pushed out and the top valve shut; the bottom valve was opened, allowing the feed to be released in the reactor. After the change, the levels of oxygen were lower than 0.6%.

The second adjustment was related to the gas seal inside the reactor. The original apparatus consisted of a metal plate attached externally at the top of the reactor lid, giving support to the motor, connecting it to the mixing blades. In the centre of the of the plate there was a metal pipe (approximately 10 cm), placed trough the lid inside the reactor, allowing the top of the mixing blade to be connected with the motor. This tube served as a gas seal, when the volume inside the reactor was higher than its base. However, due to the quality of the material and type of plate-pipe fitting, it ended up corroding and/or breaking, allowing gas to escape from inside the reactor. The problem was solved by

changing the quality of the material (stronger and longer) and the fitting (screwed to the plate).

The choice of materials to assemble bench scale anaerobic digestion reactors can have significant influence during the experiment. It was proved, however, that with simple adjustments, perceived problems can be easily mitigated.

4.2.1.2 System recovery during the start up

During the start-up two reactors were recovered after thermal shock, due to technical failure, and subsequently drop in the pH and biogas production. In the first instance, one chemostat reactor was opened to perform technical repair with its temperature probe being removed from the inside. As the electric blanket heats according to signal emitted from the probe, which was placed outside the reactor (room temperature 18 ˚C) during the repair, the blanket warmed up constantly in order to achieve the programmed mesophilic temperature. When the technical repair finished (approximately 2 hours later), the temperature inside the reactor was 62 ˚C. As a result, the methane content in the biogas and pH rapidly dropped from 50% and 7.8 to 23% and 7.4, respectively (Figure 4.1).

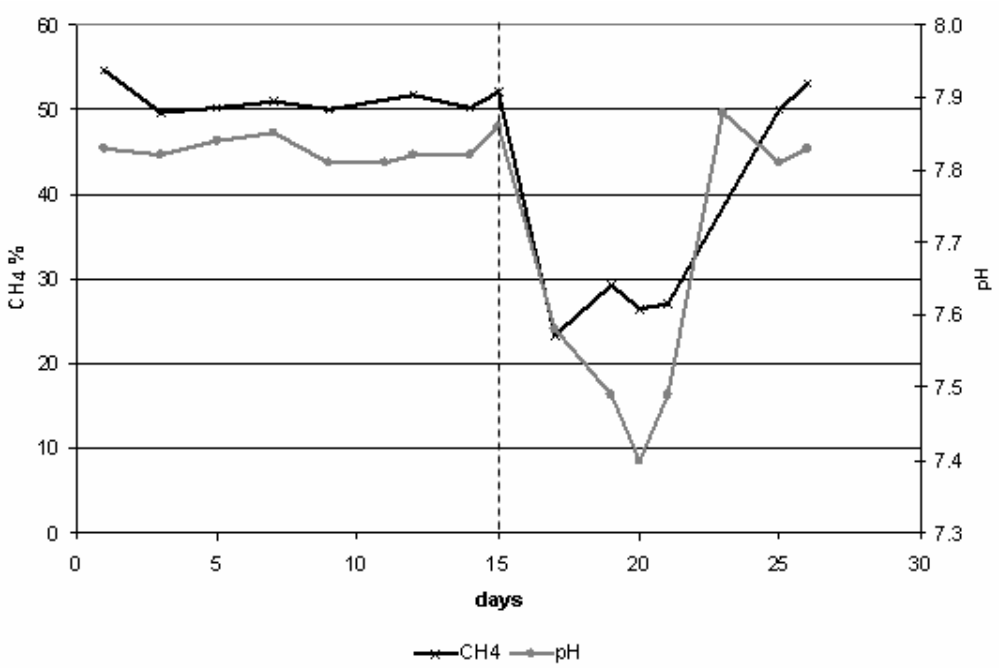

Figure 4.1 – System recovery after thermal shock (dashed line), indicating drop in the methane content and pH.

Considering that methanogens are sensitive to abrupt change in temperature, the low levels of methane recorded could be related to the reduction of these microorganisms; this suggestion was correlated to the drop in the pH level (although within the tolerable close to neutral range (Golueke, 2003), meaning acid accumulation and few methanogens to catabolise it. To mitigate the problem, the reactor was left without feed for three days, allowing time for the methanogens to start recovering as well as avoiding excessive accumulation of acids; a gradually increase in the feed (1, 1.5 and 3 gVS/L) occurred in the following days. A total recovery of the system occurred twelve days later, demonstrating resilience of the microbial community able to cope with stress, with methane content in the biogas and pH reaching 55% and 7.8, respectively; the quantity of biogas also increased.

Similar incident also occurred at the beginning of the acclimatisation process, when other reactor was heated to 52 ˚C for 3.5 hours due to failure in the temperature probe; although at that time there was no gas analyser available to measure the drop in the methane

content, it was possible to record the decrease in the pH. A controlled feed system was also performed, in order to optimise the population of methanogens, demonstrated by the increasing in the biogas production, with reactor stabilised in fifteen days.

The feasibility of recovering a digester after thermal shock where drop in the pH level and decrease in the quantity of biogas and methane content may be expected, can be of interest for anaerobic digester operators as it could be controlled through the feeding, avoiding extra costs with buffering process or even mitigating a failed reactor. Results regarding system recovery after thermal shock are limited in the literature and can be a subject for further investigation.

4.2.1.3 Bench scale trial

Monitoring the volatile solids loading rate during anaerobic digestion is important, as a constant loading rate will guarantee the stability of the system. An overload of organic material could result in excessive production of acid leading to the inhibition of methanogenic activity (Pahl *et al.*, 2008). The loading rates utilised during the trials ranged from 2.5 to 4.5 g VS/L were comparable to those reported in related studies on the co-digestion of mixed waste or source-segregated organic fraction of municipal solid waste, 0.7–4.5 g VS/L/d (Angelidaki *et al.*, 2006; Davidsson *et al.*, 2007; Gómez *et al.*, 2006; Hartmann and Ahring, 2005; Krupp *et al*., 2005; Pahl *et al*., 2008; Sosnowski *et al*., 2003; Vogt *et al*., 2002); this study recorded methane content of 51%, compared to the study with MBT material reported by Pahl *et al.* (2008) ranging from 40 to 50%.

The result for the anaerobic digestion trials indicated an overall biogas yield of 230 and 415 mL/gVS for MBT-1 and MBT-2, respectively, with a confidence interval (95%) between 235 and 360 mL/gVS for MBT-1 and between 320 and 510 mL/gVS for MBT-2. The

average methane contents, for MBT-1 and MBT-2, in all digestion trials were considered constant at 51% (Figure 4.2 and Table 4.2). Methane yield was estimated at 150 and 210 mL CH_4/gVS added, for MBT-1 and MBT-2, respectively. The mass balance analysis indicated 60% and 72% volatile solid destruction, for MBT-1 and MBT-2, respectively. The pH varied from 7.9 at the beginning of the experiment to 7.2 at the end, for both MBT-1 and MBT-2 (Figure 4.3), representing the stability of the system, efficient production of biogas and waste reduction (inferred by volatile solid destruction).

The three digestion trials presented no significant difference regarding the methane content, 51%; however, during the first trial, the biogas yield produced was slightly higher, considering the volume of gas captured in the bags. Unfortunately, at the time, the volume of biogas could not be measured, due to lack of gas flow meter on site.

The low levels of methane could be related to the quality of the waste, as MBT materials contained high concentration of recalcitrant materials such as fibre and lignin. In this experiment were also found small polystyrene sphere that, due to its floatation, tended to accumulate with other non biodegradable material, at the top of the digestate. As a measure to avoid damaging the stirring and temperature probe, the reactors were opened to remove the excess of these materials.

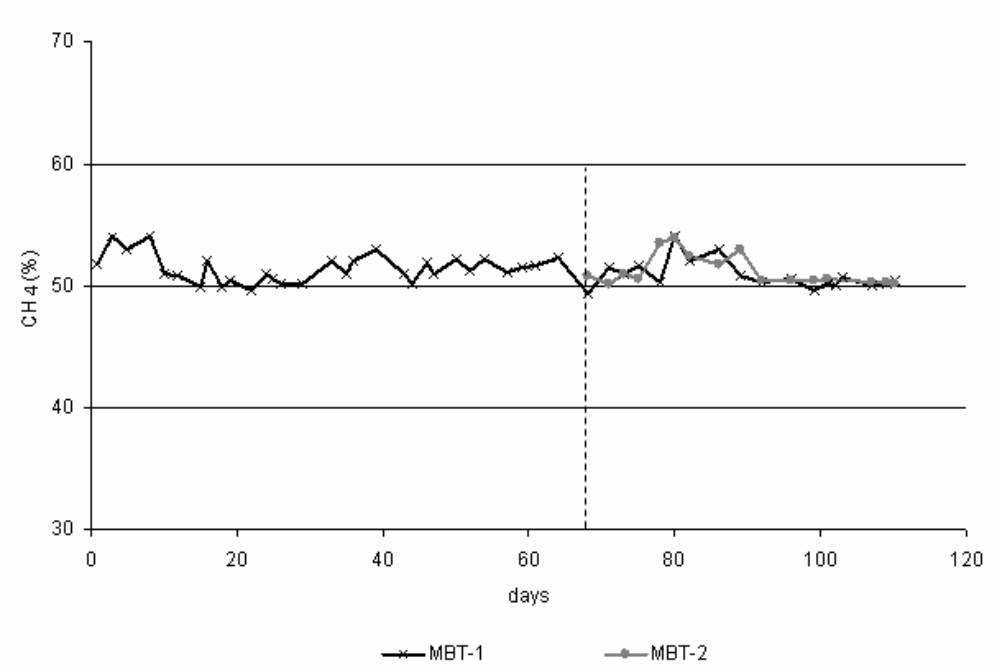

Figure 4.2 – Methane content during anaerobic digestion trials; dashed line indicates beginning of third trial, when second feedstock started to be used.

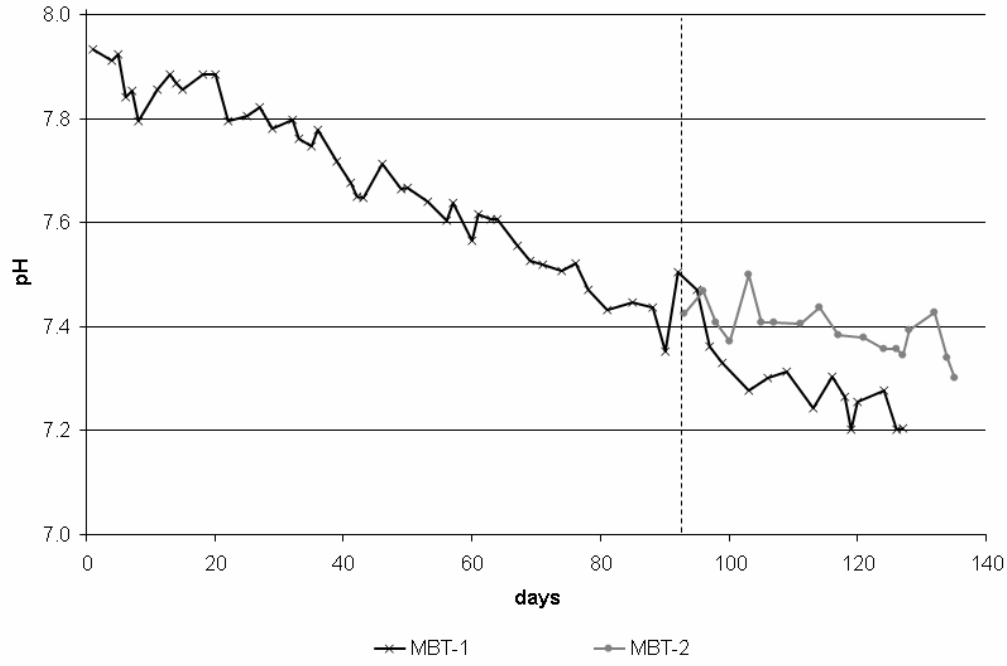

Figure 4.3 – pH range during the digestion trial; dashed line indicates beginning of third trial, when second feedstock started to be used.

Table 4.2 – Summary of methane content (%) for each digestion trial. Each value represents the mean amount of methane from triplicate samples; standard deviation is shown in parentheses.

Waste	Methane (%±sd)			
	Trial 1	Trial 2	Trial 3	Total
MBT-1	50 (1.1)	52 (1.2)	50 (1.2)	51 (1.0)
MBT-2	52 (2.8)	51 (1.2)	50 (1.8)	51 (1.0)

The biogas yield analysed through the gas flow meter was 295 ± 30 and 415 ± 4 (±sd) mL/gVS added, for MBT-1 and MBT-2, respectively. Considering the quality of biogas produced (methane content), it could be estimated a production of 150 and 210 mL CH_4/gVS added, for MBT-1 and MBT-2, respectively.

The biogas measurement also indicated an increase in the H_2S levels (Figure 4.4) when waste material MBT-2 started to be fed in the reactors, indicating the influence of the feedstock material. The levels reached the equipment maximum reading capacity of 550 ppm (mg/L) in few days, which could indicate that the actual levels during the experiment were higher. Sulfate reducing bacteria are responsible for the production of H_2S in the anaerobic digestion process, which is also inhibitory to the methanogens at low concentrations (Gerardi, 2003). As a decrease in the methane content was not observed with the introduction of the MBT-2, the inhibitory effect from the H_2S in the methanogens could be affecting the actual biogas potential.

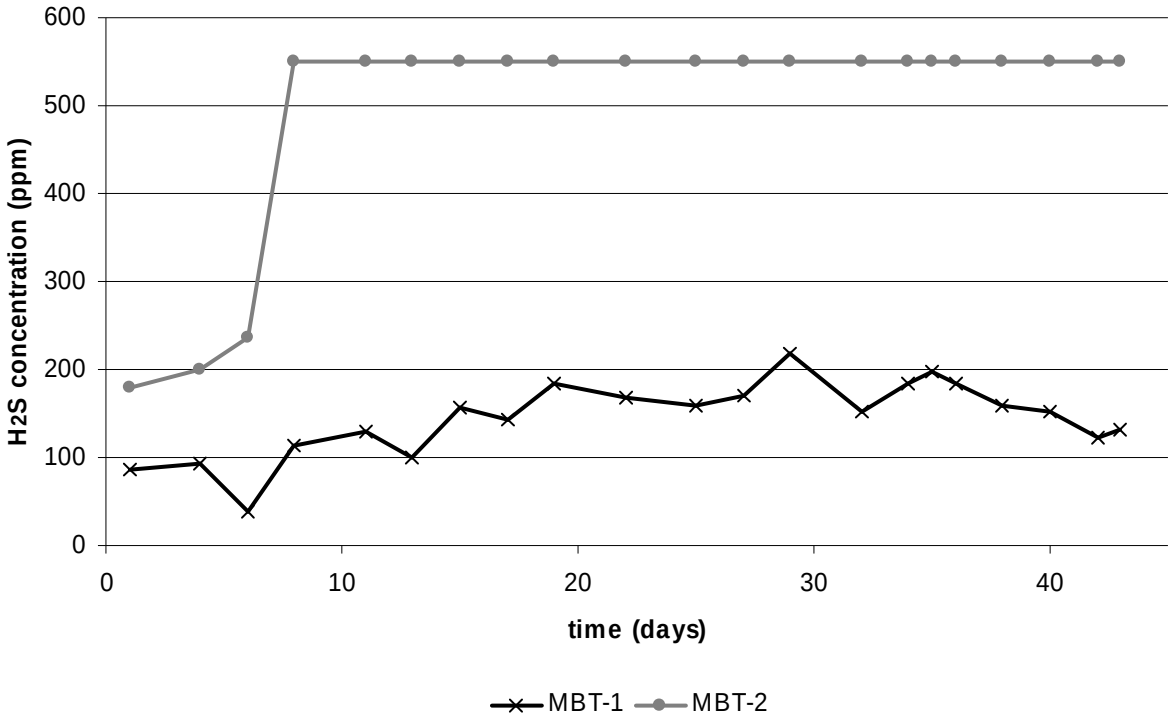

Figure 4.4 – Levels of H_2S during anaerobic digestion; peak represent limit of measurable range.

Carbon monoxide (CO) can normally be found in the biogas in trace levels (Evans, 2001; IWM, 1998; Polprasert, 1996); however, its presence is more relevant for monitoring landfill biogas as an indicator of spontaneous combustion within the buried waste. In this study, its presence were not relevant, but could also be monitored with the gas analyser. Levels of CO increased constantly, after the introduction of the MBT-2, and proportionally to the levels of H_2S, up to the maximum capacity of 1100 ppm (mg/L). According to technical information provided by the manufacturer of gas analyser (*Geotechnical Instruments, UK*), although the levels of H_2S could be considered as accurate, the high levels of CO could be explained as a result of a cross gas effect on the CO cell, in the equipment. Although the cell had an onboard filter to help against the cross gas effect from H_2S in a typical application, when applying very high levels of H_2S this would increase the sensitivity of the CO to H_2S over time, resulting in a false reading (Bush, 2008). It was also

informed that as the levels of carbon monoxide were increasing proportionally to the levels of hydrogen sulfide, the final H_2S concentration could be proportionally higher to the CO concentration, when the last reached its measurement limit of 1100 ppm (Bush, 2008). The reasons for the increase in the H_2S could be the subject for further investigation.

The presented values are comparable to other digestion trial using MBT material: Pahl *et al*. (2008) reported biogas production ranging from 130 to 240 mL with methane content from 40 to 47%. These values are also comparable to data from source segregated MSW analysis, ranging from 100 to 700 mL/gVS, reported elsewhere (Davidsson *et al*., 2007; Hartmann and Ahring, 2005; Rao and Singh, 2004; Sosnowski *et al.*, 2003, (Angelidaki *et al.*, 2006)) indicating the stability of the system and efficiency in the biogas production.

4.2.1 Batch system

The batch digestion trial was analysed for 40 days, with biogas production occurring until the day 20, when started to decrease, until no more gas was produced, reaching a plateau (Figure 4.5). The results from the batch digestion trial demonstrated volatile solids destruction of 70% for the waste MBT-1 and 74% for the waste MBT-2 (standard deviation 4 and 2, respectively). The total biogas production for the MBT-1 was 1340 mL while for MBT-2 was 1190 mL, with an average methane content of 54% for both. The methane yields were 120 and 108 mL/gVS, with a confidence interval (95%) between 70 and 150, and 35 and 180 mL/gVS, for MBT-1 and MBT-2, respectively. The final pH for MBT-1 was 7.6 and MBT-2 was 7.4, indicating biological stability in the system.

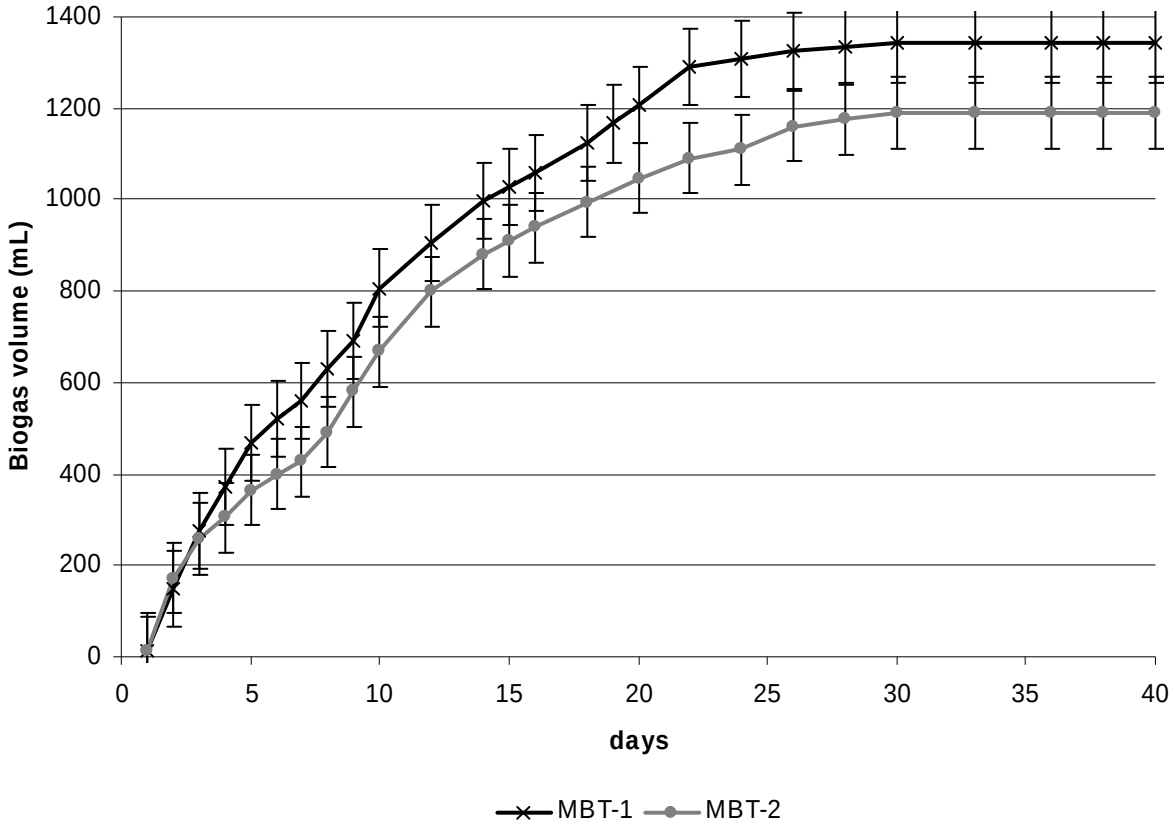

Figure 4.5 – Cumulative biogas production during batch trial; plateau reached after 33 days.

The biogas yield during the batch trial was 220 and 200 mL/gVS, with average methane content of 54% for both MBT-1 and MBT-2; the level of volatile solid destruction was 60 and 73%, respectively. The data set were comparable to values reported elsewhere for source segregated waste stream (Arsand, 2006; Davidsson *et al.*, 2007; Sosnowski *et al.*, 2003; Zhang *et al.*, 2007); the VS/TS tested by Zhang *et al*. was 83%, compared to 62% in this study, so it is believed that with similar conditions, the present experiment could achieve higher biogas yield.

4.3 Heavy metal analysis

The results from the ICP-MS analysis for the heavy metal content showed an increase in the levels of some metals for specific waste stream (Table 4.3). An accumulation of copper, nickel and zinc were observed as the trial progressed. At the end of the experiment levels of copper, nickel and zinc in MBT-1 and 2 reached between two and six order of magnitude higher than those of the initial samples (Table 4.3). In contrast levels of lead, chromium and cadmium didn't show any significant concentration changes during the experiment. Relative toxicities and absolute EC50 values, in mg/L (50% inhibition of microbiological activity) of heavy metals for anaerobic digestion have been reported (Codina *et al.*, 1998) as Zn (50) > Cr (50) > Cu (100) > Cd (200) > Ni (350). All values presented for both feedstocks are lower and reported in µg/L. The accumulation of heavy metals during the anaerobic digestion of MBT-derived material may be expected and monitoring should be placed when operating the system, as it can achieve toxic levels as well as present inhibitory effect in the microbial activity. The reason why levels of lead, chromium and cadmium did not show any significant concentration changes was not determined and could be the subject of further investigation.

Table 4.3 – Heavy metal concentration.

		Heavy metal concentration µg/L (±s.d.)		
Metal	**Waste**	**T1**	**T2**	**T3**
Zinc	MBT-2	50 (10)	52 (20)	302 (12)
Nickel	MBT-1	151 (29)	364 (307)	703 (248)
	MBT-2	171 (24)	180 (16)	435 (29)
Copper	MBT-1	25 (4)	81 (21)	180 (45)
	MBT-2	18 (1)	21 (10)	52 (23)
Lead	MBT-1	33 (18)	44 (7)	24.6 (6)
	MBT-2	44 (5)	24 (12)	29.6 (11)
Chromium	MBT-1	17(2)	21 (9)	12.7 (5)
	MBT-2	19 (11)	12 (9)	12.7 (5)
Cadmium	MBT-1	<1 (<1)	1 (<1)	<1 (<1)
	MBT-2	<1 (<1)	<1 (<1)	<1 (<1)

4.4 Microbiological community analysis – PLFA

A representative gas chromatogram of FAME distribution is presented (Figure 4.6.). A total of 34 peaks were identified, with specific PLFA emerging and disappearing during the treatment, demonstrating a complex and diverse community (Figure 4.7). No individual species were identified. The changes in reactor bacterial community structures during the waste degradation experiments were examined by PLFA analysis.

PLFA patterns analyzed from the different reactors presented a shift in the community profile when comparing two different treatments applied (Figure 4.8); however, with no statistical significance; similar result was found for bacterial communities when compared over time, p>0.05 (p=0.16 and 0.48, respectively). The microbial results demonstrated stable and dynamic system with resilient communities present in the reactors, capable to cope with environmental perturbations. Similar approach was used in studies with marine pollution (Coulon *et al.,* 2007) and epidemiologic microorganisms (Barbolla *et al*., 2002).

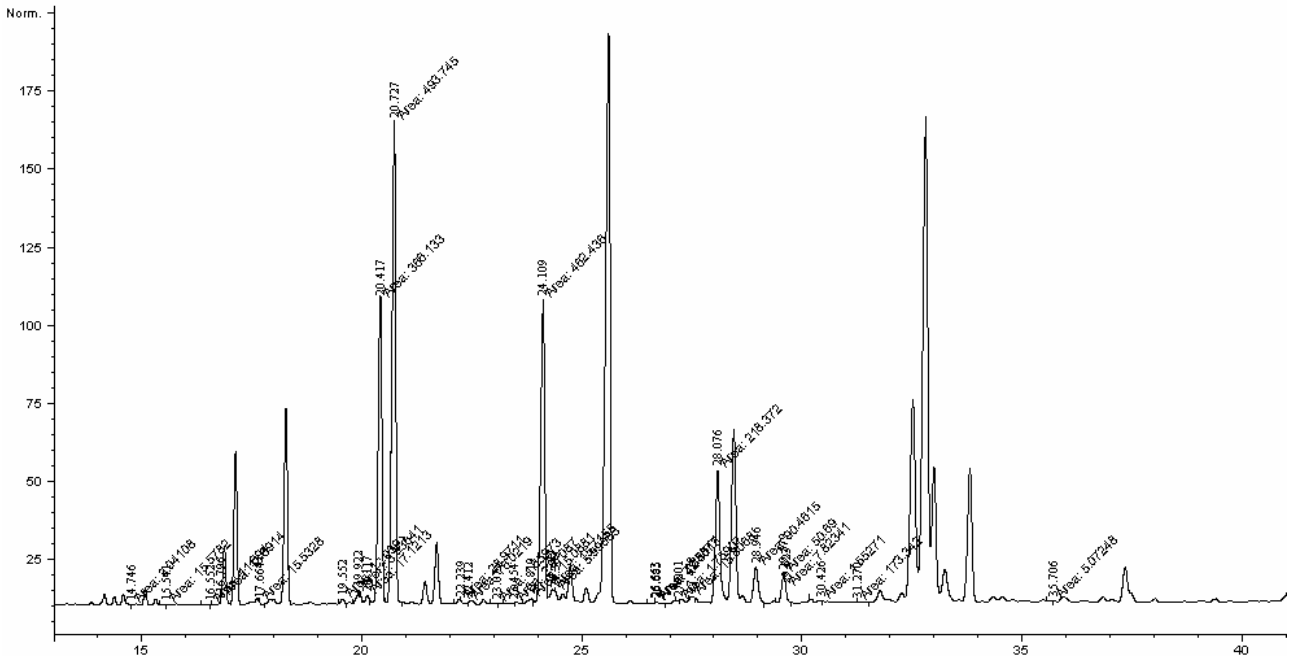

Figure 4.6 – Gas chromatogram result with FAME peaks.

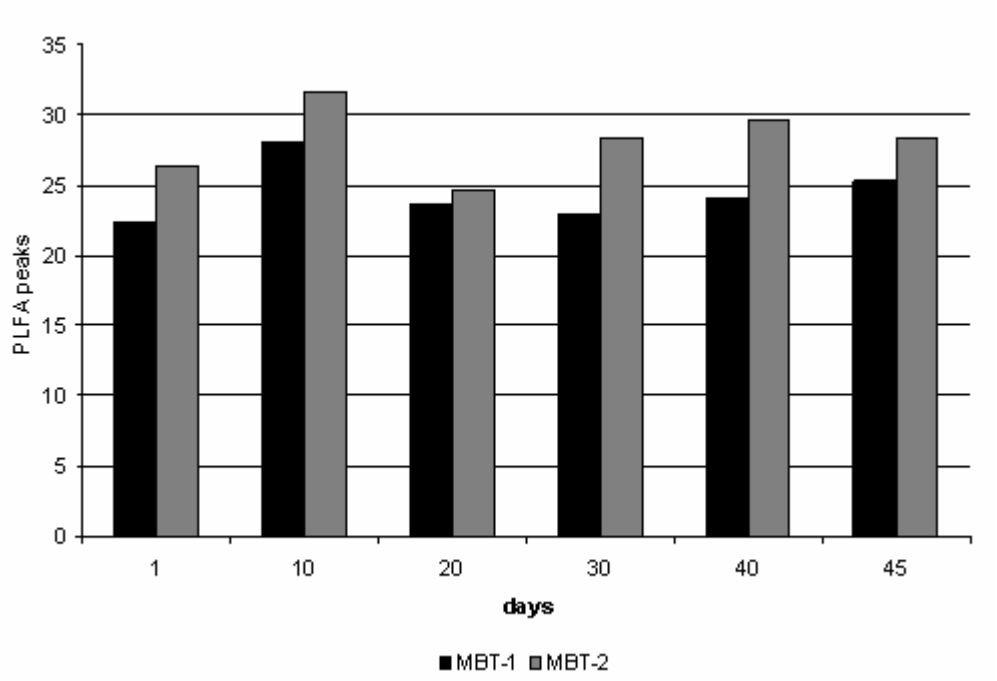

Figure 4.7 – Average PLFA peaks.

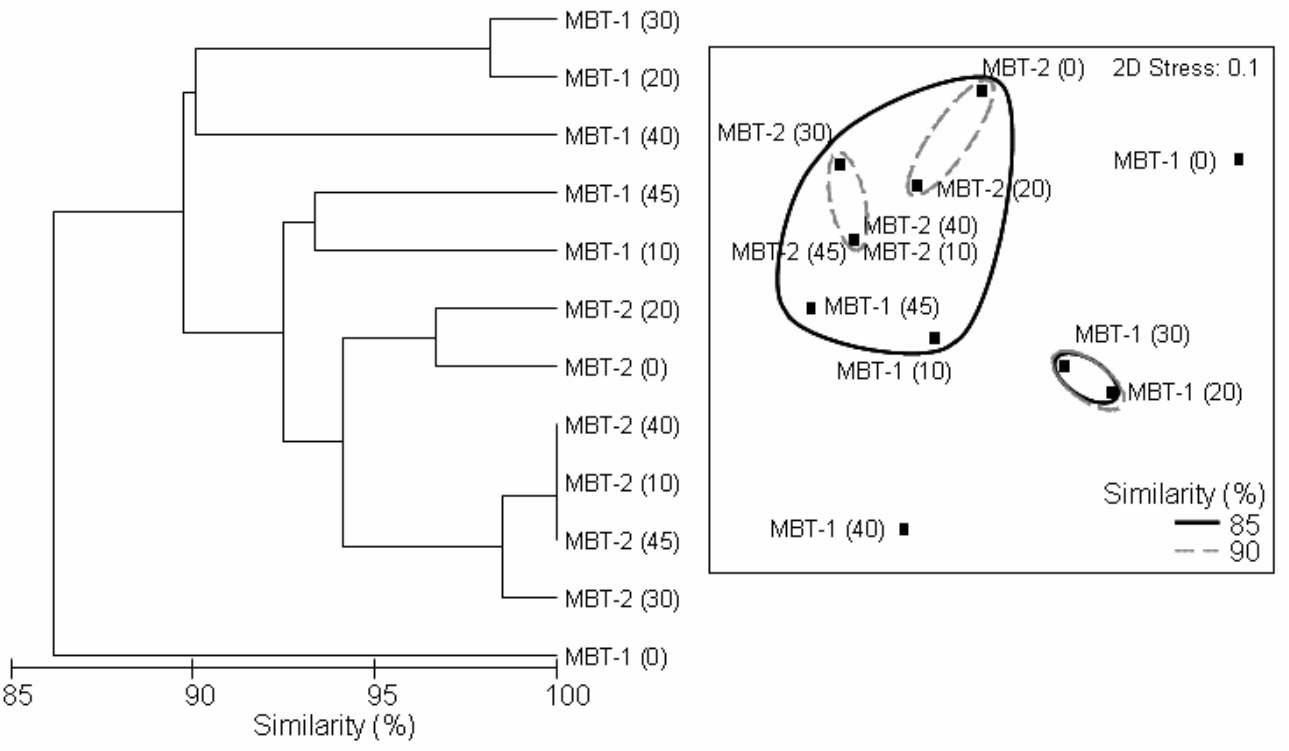

Figure 4.8 – Changes in anaerobic digestion bacterial community structures based on PLFA analysis from two MBT wastes + sampling day (parenthesis). Hierarchy cluster analysis on the left side and 2D-nMDS configuration on the right side.

4.5 Limitations

The present experiment was conducted using two different waste streams originated from two full scale MBT plants. Due to logistic to collect the samples, the material was collected in one day. This way, seasonal variation in the feedstock quality as well as variation during the week (as different communities are provided with the service in different days) could not be determined.

The levels of biogas produced during the semi-continuous trial could not be recorded from the beginning of the experiment as the gas flow meter was not provided by Summerleaze AnDigestion until near the end of the research.

CHAPTER 5

CONCLUSION

The result of the anaerobic digestion trials in semi-continuous and batch feed, using MBT material from two different full scale waste treatment plants demonstrated the potential to generate high biogas yield, as well as waste volume reduction, according to volatile solids destruction. The proposed trials, using different hydraulic retention time, loading rate and waste stream as feedstock, did not show significant difference in the biogas yield neither in the methane content, which remained stable. The levels of H_2S increased considerable, reaching levels within risk of death. The heavy metal content presented a significant increased level for zinc, nickel and copper while the levels of lead, chromium and cadmium remained stable. Although accumulation was recorded, final values were lower than toxicity levels.

The phospholipid fatty acid analysis presented a shift in the community profile when comparing two different treatments applied, however, did not show statistical significance when comparing the two treatments between each other and over the time ($p>0.05$). The findings demonstrated a stable and resilient community able to cope with different parameters and stress. The stability and resilience of the system could also be verified during the start-up, when some reactors were recovered from a thermal shock and consequently decrease in the pH and biogas production. The recovery was based on controlled feed and stable conditions were recorded after two weeks.

The experiment proved anaerobic digestion with mechanical-biological waste for MSW to be an efficient waste treatment when applied for waste streams without source segregation with considerable biogas production and methane content. Although the

quantity and quality of the biogas from anaerobic digestion with MBT was lower, due to the quality of feedstock applied, when compared to anaerobic digestion of source segregated waste, the treatment proved to be a reliable and important asset for an integrated waste management approach. Legislative drivers could play an important role in more widespread adoption of the technology by setting targets for more sustainable waste management practices and standards for insuring quality outputs such as for the digestate material.

This study was considered as groundwork in the subject as few studies were reported for anaerobic digestion with MBT with waste streams from the UK. The following aspects are recommended for further research:

- analysis of the biogas production considering seasonal variation of the waste;
- analysis of the quality of the digestate prior final use/dispose;
- analysis of the concentration of the organic matter that could be mixed or attached to reduced size material after the mechanical treatment, and for that reason, would be sent for final disposal without further treatment;
- analysis of the concentration of the heavy metals during the process, specially related to lead, chromium and cadmium, that did no presented an increase during this research;
- analysis of the reasons for the elevated concentration on the levels of H_2S during anaerobic digestion treatment;
- analysis on the system recovery after induced increase and decrease of temperature as well as other physical parameters.

REFERENCES

1774/2002 - Official Journal of the European Communities, 2002. Regulation (EC) 1774/2002 of the European Parliament and of the Council of 3rd October 2002, laying down health rules concerning animal by-products not intended for human consumption.

1999/31/EC - Official Journal of the European Communities, 1999. Council Directive 1999/31/EC on the landfill of waste.

2003/30/EC - Official Journal of the European Union, 2003. Directive 2003/30/EC of the European Parliament and of the Council of 8th May 2003 on the promotion of the use of biofuels or other renewable fuels for transport.

75/442/EEC - Official Journal of the European Communities, 2003. Council Directive 1975 on waste (75/442/EEC).

Ahring, B.K., Sandberg, M. and Angelidaki, I., 1995. Volatile fatty acids as indicators of process imbalance in anaerobic digestors Applied Microbiology and Biotechnology, 43(3), 559-565.

Ahring, B.K., Ibrahim, A. and Mladenovska, Z., 2001. Effect of temperature increase from 55 to 65 ˚C on performance and microbial population dynamics of an anaerobic reactor treating cattle manure. Water Research, 35 (10), 2446–2452.

Alker, G., 2004. Assessment of digestate from the Portagester system. [online]. Available from: http://www.bioplexltd.com/Downloads/Reports/WRc_Final_Report_uc6698.pdf

Angelidaki, I., Chen, X., Cui, J., Kaparaju, P. and Ellegaard, L., 2006. Thermophilic anaerobic digestion of source-sorted organic fraction of household municipal solid waste: Start-up procedure for continuously stirred tank reactor. Water Research, 40(14), 2621-2628.

Archer, E., Baddeley, A., Kley, A., Scwager, J. and Whiting, K., 2005. Juniper report - mechanical-biological-treatment: A guide for decision makers, process, policies and markets.

Arsand, M., 2006. Sustainable and viable anaerobic digestion systems for rural areas in the North of Scotland – Report for SDRC and Integrated Bio-Gen Ltd., Scotland.

ASSURRE, 2004. Proposed Directive on the biological treatment of biowaste. [online]. Available from: http://www.assurre.eu/uploads/documents/pub-5_en-e1a0e400-bfa0-4301-9b10-fb12ad0b8757.pdf

Balkwill, D.L., Leach, F.R., Wilson, J.T., McNabb, J.F. and White, D.C. 1988. Equivalence of microbial biomass measures based on membrane lipid and cell wall components, adenosine triphosphate, and direct counts in subsurface aquifer sediments. Microbial Ecology. 16, 73–84.

Banks, C.J. and Wang, Z., 1999. Development of a two phase anaerobic digester for the treatment of mixed abattoir wastes. Water Science and Technology. 40(1), 69-76.

Barbolla, R.E., Centrón, D., Di Martino, A., Maimone, S., Salgueira, C., Famiglietti, A., Vay, C. and Catalano, M., 2003. Identification of an epidemic carbapenem-resistant *Acinetobacter baumannii* strain at hospitals in Buenos Aires City. Diagnostic Microbiology and Infectious Disease 45, 261–264.

Barker, J.C., 2001. Methane fuel gas from livestock wastes: A summary. [online]. Available from: http://www.bae.ncsu.edu/programs/extension/publicat/wqwm/ebae071_80.html

Barth, J., 2006. Treatment of biowaste: State of the art and trends in Europe. Proceedings of the ISWA Beacon Conference: Biological Treatment of Biowaste, Perugia, Italy. 10-12 May, 2006.

Bezama, A., Aguayo, P., Konrad, O., Navia, R. and Lorber, K.E., 2007. Investigations on mechanical biological treatment of waste in south america: Towards more sustainable MSW management strategies. Waste Management, 27(2), 228-237.

Bhambhani, Y. and Singh, M., 1991. Physiological effects of hydrogen sulfide inhalation during exercise in healthy men. Journal of Applied Physiology, 71, 1872-1877.

Björklund, A., Dalemo, M., and Sonesson, U., 1999. Evaluating a municipal waste management plan using ORWARE. Journal of Cleaner Production 7, 271–280.

Blanco, E. 2008. Summerleaze AnDigestion, Cambridshire. Personal communication.

Bligh, E.G. and Dyer, W.J., 1959. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Physiology, 37**,** 911–917.

BS EN 12457-4, 2002. Characterisation of waste. Leaching. Compliance test for leaching of granular waste materials and sludges. One stage batch test at a liquid to solid ratio of 10 l/kg for materials with particle size below 10 mm (without or with size reduction).

BS EN 15002, 2006. Characterization of waste - preparation of test portions from the laboratory sample.

Braber, K., 1995. Anaerobic digestion of municipal solid waste: A modern waste disposal option on the verge of breakthrough. Biomass and Bioenergy, 9(1-5), 365-376.

British Biogen, n.d. Anaerobic digestion of farm and food processing residues - good practice guidelines. [online]. Available from: http://www.r-e-a.net/content/images/articles/adgpg.pdf

Burland, S.M. and Edwards, E.A., 1999. Anaerobic benzene biodegradation linked to nitrate reduction. Applied and Environment Microbiology, 65, 529-533.

Bush, James. 2008. Geotechnical Instrument technical support. Personal communication.

Carucci, G., Carrasco, F., Trifoni, K., Majone, M. and Beccari, M., 2005. Anaerobic digestion of food industry wastes: Effect of codigestion on methane yield. Journal of Environmental Engineering, 13(7), 1037-1045.

Cecchi F., Battistoni P., Bolzonella D. and Innocenti L., 2002. Characteristics of the organic fraction of municipal solid wastes in Europe for different sorting strategies and related performances of the anaerobic digestion process. Merida-Yucatan, Mexico. VII Latin American Symposium on Anaerobic Digestion, 22-25 October.

Cecchi, F. and Battistoni, P., 2003. Use of hydrolysis products of the OFMSW for biological nutrient removal in wastewater treatment plants, in: Mata-Alvarez, J. (Ed.), Biomethanization of the Organic Fraction of Municipal Solid Wastes. IWA Publishing, UK, 229-264.

Cecchi, F., Traverso, P., Pavan, P., Bolzanella, D. and Innocenti, L., 2003. Characteristics of the OFMSW and behaviour of the anaerobic digestion process, In: Mata-Alvarez, J. (Ed.), Biomethanization of the Organic Fraction of Municipal Solid Wastes. IWA Publishing, UK, pp. 141-199.

Chen, C., Macarie, H., Ramirez, H., Olmos, A., Ong, S.L., Monroy, O. and Liu, W., 2004. Microbial community structure in a thermophilic anaerobic hybrid reactor degrading terephthalate. Microbiology 150(10), 3429-3440.

Clarke, K.R and Warwick, R.M., 2001. Change in marine communities: An approach to statistical analysis and interpretation. 2nd Ed. PRIMER-E, Plymouth, UK, 172 pp.

Coates, J.D., Chakraborty, R., Lack, J.G., O'Connor, S.M., Cole, K.A., Bender, K.S. and Achenbach, L.A., 2001. Anaerobic benzene oxidation coupled to nitrate reduction in pure culture by two strains of *Dechloromonas*. Nature, 411, 1039-1043.

Codina, J.C., Munoz, M.A., Cazorla, F.M., Perez-Garcia, A., Morinigo, M.A. and De Vicente, A., 1998. The inhibition of methanogenic activity from anaerobic domestic sludges as a simple toxicity bioassay. Water Research, 32 (4), 1338–1342.

Collins, L.J. and Paskins, A.R., 1987. Measurement of trace concentrations of hydrogen in biogas from anaerobic digesters using an exhaled hydrogen monitor. Water Research, 21(12), 1567-1572.

Coulon, F., McKew, B.A., Osborn, A.M., McGenity, T.J., Timmis, K.N., 2007. Effects of temperature and biostimulation on oil-degrading microbial communities in temperate estuarine waters. Environmental Microbiology, 9, 177-186.

Davidsson, □, Gruvberger, C., Christensen, T.H., Hansen, T.L. and Jansen, J.l.C., 2007. Methane yield in source-sorted organic fraction of municipal solid waste. Waste Management, 27(3), 406-414.

De Baere, L., 2000. Anaerobic digestion of solid waste: State-of-the-art. Water Science and Technology, 41(3), 283-290.

Dearman, B., Marschner, P. and Bentham, R.H., 2006. Methane production and microbial community structure in single-stage batch and sequential batch systems anaerobically co-digesting food waste and biosolids. Applied Microbiology and Biotechnology, 69(5), 589-596.

DEFRA, 2005. Mechanical biological treatment & mechanical heat treatment of municipal solid waste - waste implementation programme - new technologies. [online]. Available from: http://www.milton-keynes.gov.uk/recycling/documents/8._MechanicalBiologicalTreatment.pdf.

DEFRA, 2006. Animal by-products: Treatment in approved composting or biogas plants of animal by-products and catering waste. [online]. Available from: http://www.defra.gov.uk/animalh/by-prods/wastefood/compost-qanda.htm#6.

DEFRA, 2007. Waste Strategy for England. [online]. Available from: http://www.defra.gov.uk/environment/waste/strategy/strategy07/pdf/waste07-strategy.pdf.

Demuynck, M. and Nyns, E. J. and Naveau, H. P., 1984. A review of the effects of anaerobic digestion on the odour and on disease survival, In: Gasser, J. K. R. (Ed.), Composting of agricultural and other wastes. Elsevier Applied Science, London and New York.

Deurwaarder, E.P., 2005. Overview and analysis of national reports on the EU Biofuel Directive - prospectus and barriers for 2005. Report ECN-C-05-042. [online]. Available from: http://www.ecn.nl/docs/library/report/2005/c05042.pdf.

Davidsson, A., Gruvberger, C., Christensen, T.H., Hansen, T.L. and Jansen, J. la C., 2007. Methane yield in source-sorted organic fraction of municipal solid waste. Waste Management, 27 (3), 406–414.

DFT - Department for Transport, 2004. Towards a UK strategy for biofuels. [online]. Available from: http://www.dft.gov.uk/consultations/archive/2004/tuksb/towardsaukstrategyforbiofuel2047?page=2.

DTI - Department of Trade and Industry, 2003. Energy white paper - our energy future - creating a low carbon economy. Report No Cm 5761. [online]. Available from: http://www.berr.gov.uk/files/file10719.pdf.

Edelmann, W., 2003. Products, impacts and economy of anaerobic digestion of OFMSW, In: Mata-Alvarez, J. (Ed.), Biomethanization of the organic fraction of municipal solid wastes. IWA Publishing, UK, pp. 265-301.

Edelmann, W., Schleiss, K. and Joss, A., 2000. Ecological, energetic and economic comparison of anaerobic digestion with different competing technologies to treat biogenic wastes. Water Science and Technology 41(3), 263-273.

Einola, J.M., Karhu, A.E. and Rintala, J.A., 2008. Mechanically–biologically treated municipal solid waste as a support medium for microbial methane oxidation to mitigate landfill greenhouse emissions. Waste Management, 28(1), 97-111.

Environment Agency, 2005. Guidance on monitoring MBT and other pre-treatment processes for the landfill allowances schemes (England and Wales). [online]. Available from: http://www.environment-agency.gov.uk/commondata/acrobat/new_mbt_1154981.pdf.

Eriksson, K.E.L., Blanchette, R.A. and Ander, P., 1990. Biodegradation of lignin, In: Timmel, T.E. (Ed.), Microbial and Enzymatic Degradation of Wood and Wood Components. Springer-Verlag, pp. 237-247.

ESA - Environmental Services Association, 2004. European Commission's discussion document on a Biowaste Directive. [online]. Available from: http://www.esauk.org/work/briefings/biowaste.asp.

ESA - Environmental Services Association, 2008. Possible European biowaste and biogas Directives. [online]. Available from: http://esauk.org/press/?release_id=142.

EST- Energy Saving Trust. Biogas. 2007. [online]. Available from: http://www.energysavingtrust.org.uk/fleet/Vehicles/Alternativefuels/Biogas/.

Eurostat, 2005. Europe in figures - yearbook. In: Defra - Waste Strategy for England 2007. [online]. Available from: http://www.defra.gov.uk/environment/waste/strategy/strategy07/pdf/waste07-strategy.pdf.

Evans, G., 2001. Biowaste and biological waste treatment, James & James, London.

Favoino, E., 2005. The role of MBT in EU strategies for MSW management: Trends, experiences and regulatory aspects. Proceedings of Biometa 05 - VII Jornadas sobre Biometanizacion de Residuos Solidos Urbanos. Barcelona, 2005.

Fricke, K., Santen, H. and Wallmann, R., 2005. Comparison of selected aerobic and anaerobic procedures for MSW treatment. Waste Management 25, 799–810.

Frostegård, Å, Tunlid, A. and Bååth, E., 1991. Microbial biomass measured as total lipid phosphate in soils of different organic content. Journal of Microbiological Methods, 14(3), 151-163.

Fruteau, H., 2006. Valorga international. Personal communication.

Garcia, J., Patel, B.K.C., Ollivier, B., 2000. Taxonomic, phylogenetic, and ecological diversity of methanogenic *Archaea*. Anaerobe 6(4), 205-226.

Gayat, A., 2002. Anaerobic digestion of municipal solid waste: Assessment of technical and economic viability of available systems. MSc. Thesis, Cranfield University.

Gerardi, M.H., 2003. The microbiology of anaerobic digesters, John Wiley & Sons, Inc. Somerset, NJ. 186 pp.

Golueke, C., 2002. Principles of anaerobic digestion - the BioCycle guide to anaerobic digestion, The JG Press, Pennsylvania.

Gómez, X., Cuetos, M.J., Cara, J., Morán, A. and García, A.I., 2006. Anaerobic co-digestion of primary sludge and the fruit and vegetable fraction of the municipal solid wastes: Conditions for mixing and evaluation of the organic loading rate. Renewable Energy, 31(12), 2017-2024.

Grbić-Galić, D. and Vogel, T.M., 1987. Transformation of toluene and benzene by mixed methanogenic cultures. Applied and Environmental Microbiology, 53, 254-260.

Gujer, W. and Zehnder, A.J.B., 1983. Conversion process in anaerobic digestion. Water Science and Technology, 15(8-9), 127-167.

Hartmann, H., Ahring, B.K., 2005. Anaerobic digestion of the organic fraction of municipal solid waste: influence of co-digestion with manure. Water Research 39 (8), 1543–1552.

Hawkins, R.G.P. and Shaw, H.S., 2004. The practical guide to waste management law, Thomas Telford Publishing, London.

Hill, D.T. and Bolte, J.P., 1989. Digester stress as related to iso-butyric and iso-valeric acids. Biological Wastes, 28(1), 33-37.

Hill, D.T. and Holmberg, R.D., 1988. Long chain volatile fatty acid relationships in anaerobic digestion of swine waste. Biological Wastes, 23(3), 195-214.

Houghton, J.T., Ding, Y., Griggs, D.J., Noguer, M., van der Linden, P.J., Dai, X., Maskell, K. and Johnson, C.A. (Eds.), 2001. IPCC - Climate Change 2001: The scientific basis - contribution of working group I to the third assessment report of the Intergovernmental Panel on Climate Change, Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

IEA, 2003. The utilisation of CO_2 - Zero emissions technologies for fossil fuels. [online]. Available from: http://www.iea.org/textbase/papers/2003/CO2_Util_Fossil_Fuels.pdf#search=%22iea%2Bplant%20growth%22.

IWM, 1998. Anaerobic digestion, A detailed report on the latest methods and technology for the anaerobic digestion of municipal waste.

Kasai, Y., Takahata, Y., Manefield, M. and Watanabe, K., 2006. RNA-based stable isotope probing and isolation of anaerobic benzene-degrading bacteria from gasoline-contaminated groundwater. Applied and Environmental Microbiology, 72, 3586-3592.

Kaseng, K., Ibrahim, K., Paneerselvam, S.V. and Hassan, R.S., 1992. Extracellular enzymes and acidogen profiles of a laboratory-scale two-phase anaerobic digestion system. Process Biochemistry, 27(1), 43-47.

Kirchmayr, R., Scherzer, R., Baggesen, D.L., Braun, R., Wellinger, A., 2003. Animal by-products and anaerobic digestion - requirements of the European Regulation (EC) 1774/2002. Task 37 - Energy from biogas and landfill gas. [online]. Available from: http://www.iea-biogas.net/Dokumente/IEA_ABP-Brochure_en_2.pdf.

Klass, D.L., 1984. Methane from anaerobic digestion. Science 223 (4640), 1021-1028.

Krupp, M., Schubert, J. and Widmann, R., 2005. Feasibility study for co-digestion of sewage sludge with OFMSW on two wastewater treatment plants in Germany. Waste Management, 25(4), 393-399.

Lindegaard, K., 2007. Biogas - the energy of the future? Farmers Weekly. [online]. Available from: http://www.wondertree.org.uk/pdfs/Biogas.pdf.

Lepistö, R. and Rintala , J., 1999. Kinetics and characteristics of 70 ℃, VFA-grown, UASB granular sludge. Applied Microbiology and Biotechnology, 52(5), 730-736.

Lissens, G., Vandevivere, P., Baere, L.D., Biey, E.M. and Verstraete, W., 2001. Solid waste digestors: Process performance and practice for municipal solid waste digestion. Water Science and Technology, 44(8), 91.

Liu, J., Olsson, G. and Mattiasson, B., 2004. Control of an anaerobic reactor towards maximum biogas production. Water Science and Technology, 50(11), 189-198.

Lo, K.V., Liao, P.H. and March, A.C., 1985. Thermophilic anaerobic digestion of screened dairy manure. Biomass 6, 301-315.

Lovley, D.R., Woodward, J.C. and Chapelle, F.H., 1994. Stimulated anoxic biodegradation of aromatic hydrocarbons using Fe(III) ligands. Nature 370, 128-131.

Lovley, D.R., Coates, J.D., Woodward, J.C. and Phillips, E.J.P., 1995. Benzene oxidation coupled to sulfate reduction. Applied and Environmental Microbiology, 61, 953-958.

Mackie, R.I. and Bryant, M.P., 1995. Anaerobic digestion of cattle waste at mesophilic and thermophilic temperatures. Applied Microbiology and Biotechnology, 43(2), 346-350.

Maier, R.M., Pepper, I.L. and Gerba, C.P. (Eds.), 2000. Environmental microbiology, Academic Press, London. 585 pp.

Martin, P., 2003. New and renewable energy - prospects for the 21st Century. The Renewables Obligation (amendment) Order 2003 - Statutory Consultation on late payments. URN 03/1380. [online]. Available from: http://www.berr.gov.uk/files/file22091.pdf.

Mata-Alvarez, J., 2003. Fundamentals of the anaerobic digestion process, In: Mata-Alvarez, J. (Ed.), Biomethanization of the Organic Fraction of Municipal Solid Wastes. IWA Publishing, UK, pp. 1-20.

Mata-Alvarez, J., Mace, S. and Llabres, P., 2000. Anaerobic digestion of organic solid wastes. An overview of research achievements and perspectives. Bioresource Technology, 74(1), 3-16.

McDougall, F.R., White, P.R., Franke, M. and Hindle, P., 2001. Integrated solid waste management: A lifecycle inventory, 2nd Ed. Blackwell Science, London. 544 pp.

McLanaghan, S.R.B., 2002. Delivering the Landfill Directive: The role of new & emerging technologies. [online]. Available from: http://www.cabinetoffice.gov.uk/strategy/~/media/assets/www.cabinetoffice.gov.uk/strategy/technologies%20landfill%20pdf.ashx.

Mladenovska, Z. and Ahring, B.K., 2000. Growth kinetics of thermophilic *Methanosarcina* spp. isolated from full-scale biogas plants treating animal manure. FEMS Microbiology Ecology, 31, 225–229.

Moletta, R., Escoffier, Y., Ehlinger, F., Coudert, J.P. and Leyris, J.P., 1994. On-line automatic control system for monitoring an anaerobic fluidized-bed reactor: Response to organic overload. Water Science and Technology, 30(12), 11-20.

Monnet, F., 2003. An introduction of anaerobic digestion of organic wastes - final report. [online]. Available from: http://www.remade.org.uk/Organics/organics_documents/IntroAnaerobicDigestion.pdf .

Muller, W., Fricke, K., Vogtmann, H., 1998. Biodegradation of organic matter during mechanical biological treatment of MSW. Compost Science & Utilization, 6 (3), 42–52.

Münnich, K., Mahler, C.F. and Fricke, K., 2006. Pilot project of mechanical-biological treatment of waste in Brazil. Waste Management 26(2), 150-157.

Narihiro, T. and Sekiguchi, Y., 2007. Microbial communities in anaerobic digestion processes for waste and wastewater treatment: A microbiological update. Current Opinion in Biotechnology 18(3), 273-278.

NFPA - Non-Fossil Purchasing Agency, 2004. Press release - auction of renewable obligation certificates by NFPA - results - auction again delivers record ROC prices. [online]. Available from: http://www.nfpa.co.uk/id9_info.cfm?pid=15&id=28.

NFPA - Non-Fossil Purchasing Agency, 2006. Press release: E-ROC auction of renewable obligation certificates by NFPA - results. [online]. Available from: http://www.nfpa.co.uk/id9_info.cfm?pid=15&id=50.

NGVA, 2007. Biogas - The environmentally closed loop energy system. [online]. Available from http://www.ngva.co.uk/index/fuseaction/site.articleDetail/con_id/5213.

Nordberg, Å, 2004. Review of European policies affecting implementation of anaerobic digestion. [online]. Available from: http://www.adnett.org/

Nordberg, Å, 2006. Swedish Institute of Agricultural and Environmental Engineering. Personal communication.

Oleszkiewicz, J.A. and Poggi-Varaldo, H.M., 1997. High-solids anaerobic digestion of mixed municipal and industrial waste. Journal of Environmental Engineering 123(11), 1087-1092.

Olsen, J.E. and Larsen, H.E., 1987. Bacterial decimation times in anaerobic digestions of animal slurries. Biological Wastes, 21, 153–168.

Ostrem, K., 2004. Greening waste: Anaerobic digestion for treating the organic fraction of municipal solid wastes. MSc Columbia University.

Pahl, O., Firth, A., MacLeod, I. and Baird, J., 2008. Anaerobic co-digestion of mechanically biologically treated municipal waste with primary sewage sludge – A feasibility study. Bioresource Technology, 99(9), 3354-3364.

Parawira, W., Murto, M., Read, J.S., Mattiasson, B., 2005. Profile of hydrolases and biogas production during two-stage mesophilic anaerobic digestion of solid potato waste. Process Biochem. 40(9), 2945-2952.

Pavan, P., Battistoni, P., Cecchi, F. and Mata-Alvarez, J., 2000. Two-phase anaerobic digestion of source sorted OFMSW (organic fraction of municipal solid waste): Performance and kinetic study. Water Science and Technology, 41(3), 111-118.

Plombin, C., 2003. Biogas as vehicle fuel - A European overview. Trend Setter Report No 2003:3. [online]. Available from: http://www.senternovem.nl/mmfiles/109906_tcm24-124298.pdf.

Polprasert, C., 1996. Biogas production, in: Polprasert, C. (Ed.), Organic Waste Recycling. John Wiley & Sons Ltd., Chichester, pp. 115-165.

Ponsá, S., Gea, T., Alerm, L., Cerezo, J. and Sánchez, A., 2008. Comparison of aerobic and anaerobic stability indices through a MSW biological treatment process. Waste Management, In Press, Corrected Proof.

Potts, L.G.A. and Jolly, M., n.d. Controlling and monitoring anaerobic digesters fed with thermally hydrolysed sludge. [online]. Available from: http://www.earthtech.com/documents/Controlling-monitoring_anerobic_digesters.pdf.

Rao, M.S. and Singh, S.P., 2004. Bioenergy conversion studies of organic fraction of MSW: kinetic studies and gas yield–organic loading relationships for process optimisation. Bioresource Technology 95 (2), 173–185.

REG - The Rural Enterprise Gateway Knowledge Network, 2007. Biogas & anaerobic digestion. [online]. Available from: http://www.regsw.org.uk/download.aspx?downloadfile=true&fid=3159.

Robinson, H.D., Knox, K., Bone, B.D. and Picken, A., 2005. Leachate quality from landfilled MBT waste. Waste Management, 25(4), 383-391.

Sahlstrom, L., 2003. A review of survival of pathogenic bacteria in organic waste used in biogas plants. Bioresource Technology, 87(2), 161-166.

Salminen, E. and Rintala, J., 2002/5. Anaerobic digestion of organic solid poultry slaughterhouse waste - a review. Bioresource Technology, 83(1), 13-26.

SEPA - Scottish Environment Protection Agency, 2007. Waste data digest No 7, 2005/06. [online]. Available from: http://www.sepa.org.uk/pdf/publications/wds/wdd_7.pdf

Siegrist, H., Renggli, D. and Gujer, W., 1993. Mathematical modelling of anaerobic mesophilic sewage treatment. Water Science and Technology 27(2), 25-36.

Silvey, P., Pullammanappallil, P.C., Blackall, L. and Nichols, P., 2000. Microbial ecology of the leach bed anaerobic digestion of unsorted municipal solid waste. Water Science and Technology, 41(3), 9-16.

SITA, 2004. Waste hierarchy - position paper. [online]. Available from: http://www.sita.co.uk/assets/PP_WH.pdf.

Sonakya, V., Raizada, N. and Kalia, V.C., 2001. Microbial and enzymatic improvement of anaerobic digestion of waste biomass. Biotechnology Letters, 23(18), 1463-1466.

Sosnowski, P., Wieczorek, A. and Ledakowicz, S., 2003. Anaerobic codigestion of sewage sludge and organic fraction of municipal solid wastes. Advances in Environmental Research, 7 (3), 609–616.

Soyez, K. and Plickert, S., 2002. Mechanical-biological pre-treatment of waste: State of the art and potentials of biotechnology. Acta Biotechnologica, 22(3-4), 271-284.

SSEFRA - Secretary of State for the Environment, Food and Rural Affairs, 2006. Climate Change - the UK programme 2006. CM6764. [online]. Available from: http://www.defra.gov.uk/ENVIRONMENT/climatechange/uk/ukccp/pdf/ukccp06-all.pdf.

Stanier, R.Y. (Ed.), 1989. General microbiology, 5th Ed. Macmillan Education, London. 890 pp.

Steffen, R., Szolar, O. and Braun, R., 1998. Feedstocks for anaerobic digestion. Institute of Agrobiotechnology, University of Agricultural Sciences, Viena. [online]. Available from: http://www.adnett.org/

Stegmann, R., 1996. Mechanical–biological pre-treatment before landfilling of municipal solid waste. UTA International (1), 31–35.

Sterritt, R.M. and Lester, J.N., 1980. Interactions of heavy metals with bacteria. The Science of the Total Environment, 14(1), 5-17.

Steyer, J.P., Bouvier, J.C. Conte, T., Gras, P. and Sousbie, P., 2002. Evaluation of a four year experience with a fully instrumented anaerobic digestion process. Water Science and Technology, 45(4-5), 495-502.

Switzenbaum, M.S., Giraldo-Gomez, E. and Hickey, R.F., 1990. Monitoring of the anaerobic methane fermentation process. Enzyme and Microbial Technology, 12(10), 722-730.

Thanh, N.C., 1982. Biogas technology in developing countries. A review of perspectives. In: Anozie, A.N., Layokun, S.K., Okeke, C.U., 2005. An evaluation of a batch pilot-scale digester for gas production from agricultural wastes. Energy Source Part A: Recovery, Utilization, and Environmental Effect, 27(14), 1301-1311.

Toprak, 2006. Anaerobic sludge digestion. [online]. Available from: http://web.deu.edu.tr/atiksu/toprak/ani4132.html.

Trafikkontoret - Traffic & Public Transport Authority, 2000. Biogas technology and biogas use in Sweden, 2000. In: Plobin, C. Biogas as vehicle fuel - A European overview. Trend Setter Report, 2003.

TSCTD - The Schumacher Centre for Technology and Development, n.d. Practical action - grid connection - technical brief. [online]. Available from: http://www.score.uk.com/research/Shared%20Documents/Techno-Social/grid_connection.pdf

Tunlid, A. and White, D.C. 1990. Use of lipid biomarkers in environmental samples, In: Analytical Microbiology Methods: Chromatography and Mass Spectrometry. Fox, A.; Morgan, S.L.; Larsson, L.; Odham, G. (ed.). Plenum Press, New York, p. 259–274.

UNFCCC - United Nations Framework Convention on Climate Change, 1997. Kyoto Protocol to the United Nations Framework Convention on Climate Change. [online]. Available from: http://unfccc.int/resource/docs/convkp/kpeng.pdf.

Vallini, G., Cecchi, F., Pavan, P., Pera, A., Mata-Alvarez, J. and Bassetti, A., 1993. Recovery and disposal of the organic fraction of MSW by means of combined anaerobic and aerobic biotreatments. Water Science and Technology, 27(2), 121-132.

Vandevivere, P. and De Baere, L. and Verstraete, W., 2003. Types of anaerobic digester for solid wastes, in: Mata-Alvarez, J. (Ed.), Biomethanization of the Organic Fraction of Municipal Solid Wastes. IWA Publishing, UK, pp. 111-140.

Vassilev, S.V., Braekman-Danheux, C. and Laurent, P., 1999. Characterization of refuse-derived char from municipal solid waste: 1. Phase - mineral and chemical composition. Fuel Processing Technology, 59(2-3), 95-134.

Villatoro-Monzón, W., Mesta-Howard, A. and Razo-Flores, E., 2003. (2003). anaerobic biodegradation of BTEX using Mn(IV) and Fe(III) as alternative electron acceptor. Water Science Technology, 48(6), 125-131.

Vlyssides, A.G. and Karlis, P.K., 2004. Thermal-alkaline solubilization of waste activated sludge as a pre-treatment stage for anaerobic digestion. Bioresource Technology 91(2), 201-206.

Vogt, G.M., Liu, H.W., Kennedy, K.J., Vogt, H.S., Holbein, B.E., 2002. Super blue box recycling (SUBBOR) enhanced two-stage anaerobic digestion process for recycling municipal solid waste: Laboratory pilot studies. Bioresource Technology, 85(3), 291-299.

Wang, Z. and Banks, C.J., 2003 Evaluation of a two stage anaerobic digester for the treatment of mixed abattoir wastes. Process Biochemistry, 38(9), 1267-1273.

Weelink, S.A.B., Tan, N.C.G., Broeke, H., Doesburg, W., Langenhoff, A.A.M., Gerritse, J., Stams, A.J.M., 2007. Physiological and phylogenetic characterization of a stable benzene-degrading, chlorate-reducing microbial community. FEMS Microbiology Ecology, 60, 312-321.

Weiland, P., 1993. One- and two-step anaerobic digestion of solid agroindustrial residues. Water science and Technology, 27(2), 145-151.

Wheeler, P., 2001. Biogas and more - systems and markets overview of anaerobic digestion. Task 24 - energy from biological conversion of organic waste - IEA Bioenergy. [online]. Available from: http://websrv5.sdu.dk/bio/pdf/biogas.pdf.

Wheeler, P.A. and Rome, L.D., 2002. WRC report - waste pre-treatment. A review. [online]. Available from: http://publications.environmentagency.gov.uk/PDF/SP1-344-TR-E-P.pdf.

White, D. C., 1983. Analysis of microorganisms in terms of quantity and activity in natural environments. In: Microbes in Their Natural Environment. J. Slater, R. Whittenbury, and J. Wimpenny (ed.). Cambridge University Press, New York, p. 37–66.

White, D.C., Davis, W.M., Nickels, J.S., King, J.D. and Bobbie, R.J., 1979. Determination of the sedimentary microbial biomass by extractable lipid phosphate. Oecologia. 40, 51–62.

Wiegel, J., 1990. Temperature spans for growth: Hypothesis and discussion. FEMS Microbiology Letters, 75 (2-3), 155-169.

Williams, P.T., 2005. Waste treatment and disposal, 2nd Ed. Wiley, England. 380 pp.

Zábranská, J., Stepová, J., Wachtl, R., Jenícek, P. and Dohányos, M., 2000. The activity of anaerobic biomass in thermophilic and mesophilic digesters at different loading rates. Water science and Technology, 42(9), 49-56.

Zhang, R., El-Mashad, H.M., Hartman, K., Wang, F., Liu, G., Choate, C. and Gamble, P., 2007. Characterization of food waste as feedstock for anaerobic digestion. Bioresource Technology, 98(4), 929-935.

APPENDIX A

Technical information[1]

XXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXX

Input: Municipal solid waste

Mass Flow:

- 160,000 Tpa (capacity to increase up to 220,000 Tpa).
- Metals: 7.5%
- Residue (to landfill): 23%
- Co-fuel (for cement kiln): 39%
- Organic: 27.8%
- Water: 2%

Waste is placed in a bag opener and directed through a conveyor belt to a 6.4 m Ø diameter ball mill that contains several 5.5 kg steel balls. This process reduces particles to ≤80 mm. The material goes to a trammel to separate previous material in 0-40 mm and 40-80 mm). The 40-80 mm fraction passes trough an overband magnetic separator for ferrous metal (for recycling); the residual material goes into a ballistic separator (paper, plastic and cardboard for RDF). Inert material goes through an Eddy current separator to recovery non-ferrous metals (also for recycling) and the remnants is sent to landfill. The 0-40 mm fraction (mainly putrescibles) is put through a flip-flop slotted screen which removes excess water, an air classifier and through an overband magnetic, with 5mm grid.

The organic material is then transferred to XXXXXX site, to be used in the AD plant (two stages at mesophilic temperature); before being fed, the waste is treated by wet densitometric separation to further remove plastic, glasses and ceramics.

[1] Due to commercial sensitivity, some information was removed from this document.

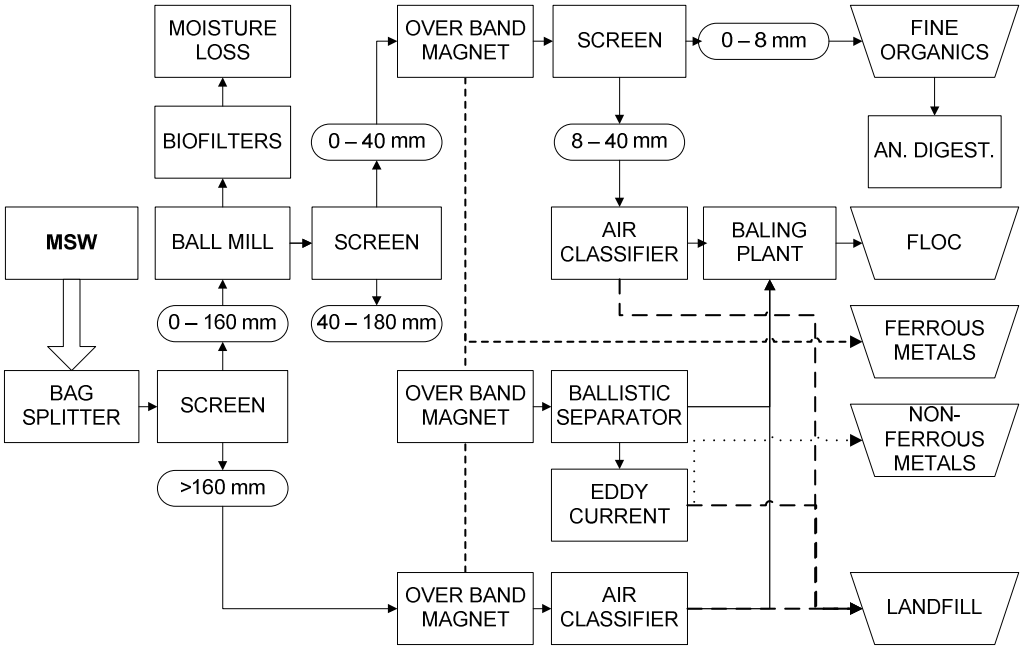

Figure A.1 – Process diagram of mechanical treatment at XXXXX site.

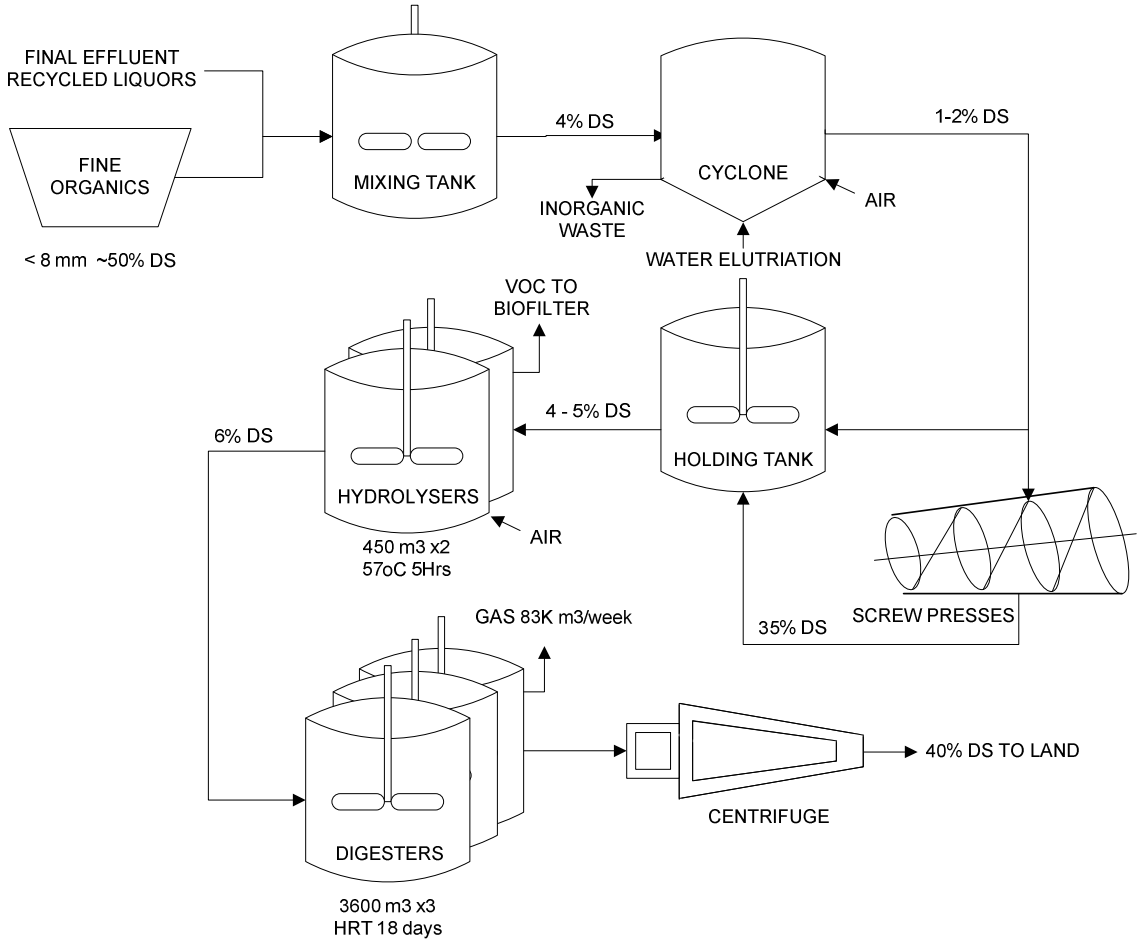

Figure A.2 – Process diagram of biological treatment at XXXXX site.

XXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXX

XXXXXXXXXXX

Input: MSW

- 7.2% Glass
- 5.4% Metals
- 22.8% Plastics
- 1.2% Non - combustible
- 63.3% BMW

Mass Flow:

110,000 Tpa

Roughly 50% Large rejects >45 mm

Roughly 50% Organic Fines <45 mm

Two stage composting of the organic fines with an assumed mass loss of around 20%.

Remaining material is roughly 50% daily cover and 50% restoration material after a screening process.

General Info:

The MSW is processed in a Rotating Dano drum which screens out the >45 mm as organic fines. Anything over 45mm is treated as large rejects. This material is then passed through an overband magnet and Eddy current separator to recover any metals. These rejects are then sent to landfill.

The organic fines are processed through a two stage composting process which aims to achieve a rough 50 / 50 split between daily cover and restoration material.

XXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXX